DOCUMENT RESUME

ED 454 661 EC 308 465

TITLE	Mental Retardation and Developmental Disabilities Branch (NICHD) Report to the NACHHD Council.
INSTITUTION	National Inst. of Child Health and Human Development (NIH), Bethesda, MD. Mental Retardation and Developmental Disabilities Branch.
PUB DATE	2001-01-00
NOTE	58p.
AVAILABLE FROM	NICHD Clearinghouse, P.O. Box 3006, Rockville, MD 20847. Tel: 800-370-2943 (Toll Free).
PUB TYPE	Reports - Descriptive (141)
EDRS PRICE	MF01/PC03 Plus Postage.
DESCRIPTORS	Adults; Autism; Children; *Clinical Diagnosis; Etiology; Federal Programs; Financial Support; Genetics; Infants; Medical Research; *Mental Retardation; Nutrition; Prenatal Care; *Prenatal Influences; *Research and Development Centers; *Research Projects; Scientific Research; Toddlers

ABSTRACT

This report highlights some of the projects supported by the Mental Retardation and Developmental Disabilities (MRDD) Branch of the Center for Research for Mothers and Children at the National Institute of Child Health and Human Development since its last report in January 1997. The MRDD Branch provides support for research, research training, conferences, and workshops on the biomedical, behavioral, and bio-behavioral aspects of mental retardation and other forms of developmental disabilities. It employs the following mechanisms to support research in the biological, behavioral, and social processes implicated in the understanding, prevention, and treatment of mental retardation and developmental disabilities: individual research grants, program projects, core support for the Mental Retardation Research Centers (MRRCs), contracts, training grants, and conference grants. The report describes the activities of the individual MRRCs and research activities addressing chromosome abnormalities, prenatal malnutrition, maternal phenylketonuria, family functioning, autism, and prenatal diagnosis. The report also includes trends of support for the MRDD branch for the past five fiscal years. The level of support of $105.1 million provided by the Branch in fiscal year 2000 represents an increase of 163 percent, compared to $64.4 million in fiscal year 1996. Appendices list sponsored conferences and workshops. (CR)

Reproductions supplied by EDRS are the best that can be made
from the original document.

EC

Mental Retardation and Developmental Disabilities Branch, NICHD

ED 454 661

EC 308405

BEST COPY AVAILABLE

2

Report to the NACHHD Council
January 2001

U.S. DEPARTMENT OF EDUCATION
Office of Educational Research and Improvement
EDUCATIONAL RESOURCES INFORMATION CENTER (ERIC)
☑ This document has been reproduced as received from the person or organization originating it.
☐ Minor changes have been made to improve reproduction quality.
• Points of view or opinions stated in this document do not necessarily represent official OERI position or policy.

Cover Illustration: Lifecycle of a Child with Phenylketonuria (PKU)
Original artwork by: Al Laoang, Medical Arts and Photography Branch, NIH, 2000

The cover illustration depicts several developmental phases of a patient with PKU, one of the many disorders on which the Mental Retardation and Developmental Disabilities (MRDD) Branch provides research support. If left untreated, this inherited metabolic disorder causes mental retardation. Advances in screening, diagnosis, and treatment enable children with PKU to develop normally and live normal, healthy lives. The MRDD Branch initiated and co-sponsored the NIH Consensus Development Conference, Phenylketonuria: Screening and Management, in October 2000, to review the current state of knowledge on PKU and Maternal PKU (MPKU) and identify directions for further PKU research.

ACKNOWLEDGMENTS

Staff members of the Mental Retardation and Developmental Disabilities (MRDD) Branch are grateful to Ms. Darlene Levenson, Chief, Referral and Program Analysis Branch, Office of Science Policy, Analysis and Communication and her staff for providing statistical information, tables, and graphs; Ms. Christina Stile of the Public Information and Communications Branch; and especially Ms. Barbara Alvarado of MRDD for collecting, collating, and organizing various segments of this document into a coherent report.

TABLE OF CONTENTS

EXECUTIVE SUMMARY 1

MENTAL RETARDATION RESEARCH CENTERS (MRRCs) 2

THE MRRC AT THE ROSE F. KENNEDY CENTER, ALBERT EINSTEIN COLLEGE OF MEDICINE, YESHIVA UNIVERSITY, BRONX, NY 4
THE MRRC AT CHILDREN'S HOSPITAL, BOSTON, MA 7
THE MRRC AT THE UNIVERSITY OF CALIFORNIA (UCLA), LOS ANGELES, CA 8
THE MRRC AT THE UNIVERSITY OF WASHINGTON (UWA), SEATTLE, WA 10
THE MRRC AT THE UNIVERSITY OF NORTH CAROLINA (UNC), CHAPEL HILL, NC 11
THE MRRC AT THE JOHN F. KENNEDY CENTER, VANDERBILT UNIVERSITY, NASHVILLE, TN 13
THE MRRC AT THE WAISMAN CENTER, UNIVERSITY OF WISCONSIN, MADISON, WI 14
THE MRRC AT THE EUNICE KENNEDY SHRIVER (EKS) CENTER, WALTHAM, MA 15
THE MRRC AT THE UNIVERSITY OF KANSAS (UKS), LAWRENCE, KS 17
THE MRRC AT THE UNIVERSITY OF COLORADO (UCO), DENVER, CO 19
THE MRRC AT THE BAYLOR COLLEGE OF MEDICINE, HOUSTON, TX 22
THE MRRC AT THE KENNEDY KRIEGER INSTITUTE, BALTIMORE, MD 23
THE MRRC AT THE UNIVERSITY OF PENNSYLVANIA (UPA), PHILADELPHIA, PA 24
THE MRRC AT THE UNIVERSITY OF CALIFORNIA (UCI), IRVINE, CA 25
THE MRRC AT THE UNIVERSITY OF ALABAMA (UAB), BIRMINGHAM, AL 25

CHROMOSOME ABNORMALITIES 26

WILLIAMS SYNDROME (WMS) 27
DOWN SYNDROME 28
X CHROMOSOME DISORDERS 29

PRENATAL MALNUTRITION 33

MATERNAL PHENYLKETONURIA (MPKU) 34

FAMILY FUNCTIONING 36

AUTISM 37

NETWORK ON THE NEUROBIOLOGY AND GENETICS OF AUTISM: COLLABORATIVE PROGRAMS OF EXCELLENCE IN AUTISM (CPEAs) 38

PRENATAL DIAGNOSIS 41

EARLY AMNIOCENTESIS VS. LATE TRANSBDOMINAL CVS (THE EATA TRIAL) 41
THE NICHD FETAL CELL STUDY (NIFTY) 42
FIRST AND SECOND TRIMESTER EVALUATION OF ANEUPLOIDY RISK (FASTER) 43

APPENDIX 1: MRDD BRANCH ACTIVITIES A-1

APPENDIX 2: MRDD BRANCH PERSONNEL A-3

FIGURES AND TABLES ..... FIGURES AND TABLES-1

EXECUTIVE SUMMARY

The Mental Retardation and Developmental Disabilities (MRDD) Branch is one of six Branches of the Center for Research for Mothers and Children at the National Institute of Child Health and Human Development (NICHD). It provides support for research, research training, conferences, and workshops on the biomedical, behavioral, and bio-behavioral aspects of mental retardation and other forms of developmental disabilities. This report highlights some of the research accomplishments made by Branch investigators, as well as other programmatic activities and future plans of the Branch, since its last presentation to the Advisory Council in January 1997.

"Mental retardation refers to substantial limitations in present functioning. It is characterized by significantly subaverage intellectual functioning, existing concurrently with related limitations in two or more of the following applicable adaptive skill areas: communication, self-care, home living, social skills, community use, self-direction, health and safety, functional academics, leisure, and work. Mental retardation manifests before age 18.

The following four assumptions are essential to the application of the definition:

- Valid assessment considers cultural and linguistic diversity as well as differences in communication and behavioral factors;
- The existence of limitations in adaptive skills occurs within the context of community environments typical of the individual's age peers and is indexed to the person's individualized needs for supports;
- Specific adaptive limitations often coexist with strengths in other adaptive skills or other personal capabilities; and
- With appropriate supports over a sustained period, the life functioning of the person with mental retardation will generally improve."

Source: *Mental Retardation - Definition, Classification, and Systems of Support, 9th Edition*, American Association on Mental Retardation, Washington, D. C., 180 pp., 1992.

The NICHD, mainly through its MRDD Branch, has the primary responsibility for providing support for research and research training in mental retardation and other forms of developmental disabilities. However, other federal agencies, several other Institutes within The National Institutes of Health (NIH), other Branches within NICHD, and the NICHD intramural research program also support and conduct research that is relevant to mental retardation and developmental disabilities.

The Branch employs several mechanisms to support research in the biological, behavioral, and social processes implicated in the understanding, prevention, and treatment of mental retardation and developmental disabilities: individual research grants, program projects, core support for the Mental Retardation Research Centers

(MRRCs), contracts, training grants, and conference grants (Table 1 and Figure 1). The relatively high proportion of investigator-initiated grants using the program project (P01) mechanism reinforces the need for multidisciplinary approaches in investigating the multifaceted aspects of mental retardation and developmental disabilities.

This report highlights some of the projects supported by the MRDD Branch. The scope of the research topics covered by the MRDD Branch is broad and rich in its diversity. Research in mental retardation and developmental disabilities draws most heavily from the fields of neuroscience, genetics, developmental neurobiology, neuroimaging, and behavioral and social sciences. It encompasses etiology, pathogenesis, prevention, and treatment of conditions that give rise to mental retardation and developmental disabilities. Recent advances are characterized by an increasing integration of behavioral and biological approaches, especially with respect to such issues as the structural basis of brain function and behavior, the interplay of organic and environmental factors, and characterization of animal models. Figure 2 summarizes the relative amounts of biomedical, behavioral, and bio-behavioral projects supported by the MRDD Branch. Appendix 1 at the conclusion of this report lists conferences and workshops that were instrumental in emphasizing some of the gaps in our scientific knowledge, research needs, and potentially fruitful research strategies that could help improve understanding of mental retardation and developmental disabilities.

This report also includes trends of support provided by the MRDD Branch for the past five fiscal years. The level of support of $105.1 million provided by the Branch in fiscal year 2000, represents an increase of 163 percent, compared to $64.4 million in fiscal year 1996 (Figure 3).

MENTAL RETARDATION RESEARCH CENTERS (MRRCs)

A unique resource of the MRDD Branch is the network of 14 MRRCs. The primary objective of these centers is to provide support and facilities for cohesive, multidisciplinary programs of research and research training in mental retardation and related aspects of human development. The MRRCs provide greater visibility for research in mental retardation within the larger research community. They successfully attract basic and clinical research scientists to address the multifaceted programs in mental retardation and developmental disabilities. Core grants both facilitate program coordination and support central research facilities. Funds for specific research projects that use these core facilities come from independent sources including the NICHD, other NIH institutes, other federal agencies, state governments, and private foundations.

Public Law 88-164, the *Mental Retardation Facilities and Community Mental Health Centers Construction Act of 1963*, authorized grants for the construction of centers for research on mental retardation and related aspects of human development. The Act stipulated that the facility would be approved if "...the application contains or is supported by reasonable assurances that: (A) for not less than 20 years after completion of construction, the facility will be used for research, or research and related purposes, for which it was constructed; (B) sufficient funds will be available for meeting the non-federal share of the cost of constructing the facility."

Following the NIH dual review system, 12 MRRCs were awarded under the provision of the Act. In 1987, the decision was made to open the competition, soliciting applications from any institution possessing the requisite programs for a center, including those that had already fulfilled their 20-year commitment. In 1991, Congress added $1.5 million to the NICHD's appropriation bill to fund two additional centers; in 1994, Congress instructed the NICHD to create a Developmental Disabilities Prevention Research Center within a university-affiliated program (UAP). Recently, this specialized center at the University of Alabama, Birmingham successfully recompeted to become an MRRC (Figure 4).

The MRRCs form the nation's major research effort for investigating the problems of mental retardation and other types of developmental disabilities. Although a significant portion of the research portfolios at the Centers consists of basic studies that are fundamental to an understanding of the biological and behavioral processes in animal models and human subjects, each MRRC directs considerable attention toward seeking solutions to practical issues and problems. Research in biological and behavioral processes areas, both basic and applied, is made possible by the vigorous outreach activities of Center scientists, administrators, and communication specialists who are involved in community education, health, and social service systems. The close working relationships between the MRRCs, UAPs, and other public and private facilities facilitates access to subject populations, and development and evaluation of enriched environmental settings. The scope of the research programs conducted at the MRRCs encompasses every known major dimension of mental retardation. Table 2 lists some of the diseases and syndromes that MRDD Branch-supported investigators are addressing. Brief highlights from the MRRCs are presented below.

THE MRRC AT THE ROSE F. KENNEDY CENTER, ALBERT EINSTEIN COLLEGE OF MEDICINE, YESHIVA UNIVERSITY, BRONX, NY

Investigators at this MRRC have a three-decade long commitment to "translating" basic science findings into clinically relevant studies and, ultimately, to possible intervention strategies. The interdisciplinary programs of this Center are organized around five main themes: 1) normal and abnormal neural development; 2) factors influencing chemical and electrical coupling of neurons; 3) systems and cognitive neuroscience; 4) neurobehavioral assessment of development in children; and 5) pathobiology of developmental and degenerative brain disorders, including possible neuroprotection and repair.

Normal and Abnormal Neural Development

Investigators in the normal and abnormal neural development group explore factors that regulate the birth of new neurons and glia as well as their specification into defined cell types. This group also studies the ongoing molecular balance between cell division and programmed cell death and the molecular events underlying dendritogenesis and axonal pathfinding in developing neurons. Investigators use both *in vivo* and *in vitro* models to probe the molecular mechanisms that regulate cell lineage decisions and cell-cell interactions during development. Some investigators study the role of ephrins and their receptors in axonal pathfinding, while others examine the roles of semaphorins and netrins in neurogenesis and axon targeting. Still others investigate the neurogenetic and cognitive features of velo-cardio-facial syndrome (VCFS), which results from the deletion of the q11 region of Chromosome 22. Investigators also focus on the etiology of mental retardation as observed in lysosomal storage diseases, where aberrant and overly exuberant dendrite formation is maintained in the adult. Researchers interested in Fragile X syndrome (FXS) examine factors that control binding proteins that in turn regulate the delivery of messenger RNA (mRNA) species to subcellular loci.

Factors Influencing Chemical and Electrical Coupling of Neurons

Investigators in the group that studies factors influencing chemical and electrical coupling of neurons focus on the plasticity of synapses and the organization of small neural networks. These studies examine short-term phenomena, like depression and facilitation, as well as long-term phenomena, like long-term potentiation and depression. Other researchers study cellular mechanisms, such as the types of synapses that mediate long- and short-term phenomena, both chemically and electrically. Both types of synapses help to trigger intracellular cascades through n-methyl-d-aspartate (NMDA) receptors, although different cascades may also be involved. Investigators also study nitric oxide-mediated long-term regulation of synaptic strength and of long term depression. Other investigators who study glutamatergic neurotransmission examine different subtypes of glutamate receptors and their regulation during nervous system development and in neurodegenerative disease, while others identify new receptors subtypes and splice variants. Other researchers address the kindling of seizures using a combination of intracranial electrical and chemical stimulations, electroencephalography

(EEG) recordings, receptor binding assays, deoxyglucose and glucose autoradiography, and *in situ* hybridization.

Investigators at the Rose Kennedy Center include a formidable group of scientists who study gap junctions. By combining molecular genetics with biophysical and biochemical techniques, these researchers characterize the molecular basis of channel gating at the single channel level. The group includes investigators who study electrophysiological and morphological features of identified electrotonic synapses *in vivo*. These studies have established links between diseases and mutations in genes that encode the connexin subunits that compose the gap junctions. The researchers found that the activity-dependent modulation of gap junctions that couple neurons electrically may be affected in seizure disorders, where alterations could influence both synchronization of neural activity and dissipation of excitatory actions of potassium.

Systems and Cognitive Neuroscience

Investigators in the systems and cognitive neuroscience group often study experimental animal models in parallel with human infants and children, including those with developmental disabilities. This research examines the pattern and sequence of events within distributed neural networks, that is, networks that often involve multiple brain regions. Investigators in this group appreciate the critical need to compare objective measures of neural function with behavioral observations. To make the comparison, the investigators outline the distribution and sequence of neocortical activation associated with sensory, motor, and cognitive processes. Studies include neurophysiological analyses at the cellular level and evaluation of scalp-recorded, event-related potentials (ERPs) in humans. In animal systems, investigators measure activity patterns in multiple laminae and in multiple cortical areas simultaneously; they correlate these activity patterns with specific brain responses using a combination of recording and drug-infusion procedures. Investigators use a high-density array of scalp recording electrodes to estimate the location, strength, and vector of intracranial generators, and to co-register brains that demonstrate activation as indexed by functional magnetic resonance imaging (fMRI). Researchers also study neural dysfunction in a variety of disease models, including neurotoxic insult, exposure to cytokines, and, most recently, genetic mutations.

Investigators who evaluate language and language disorders explore both normal and aberrant development of hearing, listening, and, most critically, language. Studies of tonotopic processing in the auditory system have outlined physiological correlates of specific components of language processing. Other researchers follow the flow of information processing both within and across cortical regions. Investigators recently extended their studies of brain response in adults and in normally developing and language-impaired children to include children with autism.

Neurobehavioral Assessment of Children

The group involved in studies of the neurobehavioral assessment of children explores normal and abnormal development of sensory, perceptual, linguistic, and cognitive functions in infancy and childhood. The group also evaluates effective interventions for developmental disorders, using observation, neurobehavioral approaches, and electrophysiological techniques. These researchers perform a number of functions: describe and analyze the development of basic sensory and cognitive domains; identify and characterize early manifestations of developmental and behavioral disorders; explore brain processes that underlie normal and deviant sensory and cognitive development; and implement and evaluate intervention strategies for an array of developmental disorders. Interests of these investigators include Specific Language Impairment (SLI), the nature of phonological deficits in children with reading disabilities, the development of language in hearing-impaired children, and possible impairments of selective attention in children with autism.

A group of investigators at the Rose Kennedy Center studies the effects of temporary hearing loss from otitis media on children's auditory and language processing, working memory, and academic achievement in early elementary grades. Longitudinal studies of preterm infants document the predictive value of early measures of recognition memory and cross-modal transfer for later more complex cognitive and language functions. Additional studies of inner-city toddlers who were exposed to lead and treated for iron deficiency attempt to increase their cognitive performance. Other investigators in this group examine individuals at the other end of the age spectrum, conducting work on memory impairment during normal aging and in dementia.

Pathobiology of Developmental and Degenerative Brain Disorders

The group involved in studies of the pathobiology of developmental and degenerative brain disorders include investigators who have conducted seminal studies on the clinical, neurochemical, enzymatic, and cellular abnormalities in Tay-Sachs and other lysosomal storage diseases. Some of these investigators are currently examining intracortical electrophysiological measures in experimental models of Batten disease. Other researchers have achieved successful metabolic correction of α mannosidosis using bone marrow transplantation and have ameliorated the neurological decline in children with Niemann-Pick Type C disease through pharmacological therapy.

Devising replacement therapies is the goal of a number of Rose Kennedy Center investigators who are interested in late-onset neural dysfunction and cell death in diseases like Alzheimer, Parkinson, Huntington, and Batten, as well as Friedreich's Ataxia and amyotrophic lateral sclerosis (ALS). The fusion of skills among Center researchers in neural development and stem cell research makes more likely the use of immature neural or glial cells for targeting affected cellular elements as a future therapy for such disorders. The rational design of neuroregenerative therapeutics requires an understanding of the differences between developmental regulation of genes and re-

expression of the same genes in the adult so that these differences can be exploited. Other investigators are exploring the influence of hormones on brain function and their possible neuroprotective roles in certain neurodegenerative disorders. These studies examine the roles of various neurotrophic factors and cytokines in preventing apoptosis and neurodegeneration or the use of trophic factors to prevent or ameliorate stroke-induced neural injury. Other researchers are examining the effects of the re-expression of developmentally regulated genes for the process of remyelination following injury. These studies are to identify and target new genes that promote axonal regeneration to the injured spinal cord to promote central axonal regeneration.

THE MRRC AT CHILDREN'S HOSPITAL, BOSTON, MA

Investigators at this MRRC bring together over 25 years of collaboration of clinical and basic science studies. One group of clinical investigators studies the interaction of cerebral hemodynamics and oxygenation in premature infants who are at high risk for brain injury, especially periventricular hemorrhage, infarction, or periventricular leukomalacia. These researchers interface with investigators involved in multi-center neuroepidemiological analyses of the pathogenetic correlates of periventricular leukomalacia. Basic researchers in this group investigate mechanisms of oligodendrocyte cell death in tissue culture and investigate the pathogenesis of white matter injury in a hypoxic-ischemic animal model. These investigators are also working to characterize oligodendroglial development in the human brain during the time of predilection for the occurrence of periventricular leukomalacia by immunocytochemistry.

Other teams of investigators focus their research on the causes and prevention of brain injury in infants with congenital heart disease who undergo cardiac surgery under deep hypothermia as well as cardiopulmonary bypass. Clinical investigators in this group study cerebral mitochondrial oxygenation and cerebral hemodynamics, electrical and chemical seizures, and cognitive and neurological outcomes in infants who undergo deep hypothermic cardiac surgery. These researchers study mechanisms of ischemic and excitotoxic neuronal death in cell culture, the neuropathological aspects of the topographic and cytological characteristics of brain injury in human infants, and relevant *in vivo* models of hypoxic-ischemic brain injury.

Another group of investigators focuses on clinical and basic studies of dyslexia. Clinical researchers in this group are working to define the neurophysiological substrates of deficits in rapid sensory processing and the impairments of temporal resolution in affected patients. Basic researchers address the neuroanatomical and neurophysiological correlates of these deficits.

Two groups of investigators that recently joined the MRRC focus on the early events involved in patterning, migration, and cellular positioning in the developing vertebrate central nervous system. The first group studies the molecular basis of vertebrate neural induction, particularly the role of the *Wnt* family of signal transducers and their receptors in axial and neural development. These researchers identified cis regulatory elements and trans acting factors that control neural gene expression and developed a novel functional expression screen for genes that regulate early neural induction both positively and negatively.

The second group studies the cellular and molecular mechanisms of neuronal migration in the developing cerebrum. These researchers found that neuronal precursors migrating over long distances to their remote destinations may begin as clones of cells that become dispersed within the cortex. Chains of cells migrate tangentially through tubes of astrocytic cells to accomplish this dispersal. Surface molecules on these glial cells induce the movement of neuronal precursors, which help to initiate neuronal migration. Secreted glial-derived proteins then determine and modulate the process. This group has done extensive work on one particular factor, astrocyte-derived Migration-Inducing Activity (MIA), which may initiate, enhance, or maintain the movement of neuronal precursors, while other molecules, such as Slit2 and Netrin, may contribute to the guidance of this activity.

THE MRRC AT THE UNIVERSITY OF CALIFORNIA (UCLA), LOS ANGELES, CA

The scope of research at the UCLA MRRC is extensive, ranging from the molecular and genetic basis of mental retardation and developmental disabilities to the socio-cultural adaptation of individuals with mental retardation and developmental disabilities and their families. This Center has made important strides in fundamental, as well as patient-based, research over the past 30 years. These studies often provide a translational link between the laboratory efforts of the Center and the clinical needs of mental retardation and developmental disabilities patients.

Investigators at the Center continue to be leaders in studies of glial cell biology and myelination. Several researchers have made significant contributions to understanding the role of myelin genes mutations in white matter disorders. UCLA MRRC investigators also discovered the *Golli-MBP* gene, a gene critical for cerebral cortical development, and identified its ramifications in immune system contributions to the pathogenesis of demyelination. UCLA investigators are studying the role of cholesterol, fatty acids, and ketone bodies in the nervous system's development of energy metabolism and the perturbations of metabolic processes by adverse environments such as alcohol, changes in iron availability, and oxygen deprivation.

Investigators who study the origin of cholesterol and fatty acids in the brain have identified local sites and novel selective transporters specific for essential fatty acids. This group also identified stage-specific genes in the oligodendrocyte lineage as well as novel effects of neurotransmitters, such as dopamine, on oligodendrocyte differentiation and myelin production. Investigators studying cell culture models for the *in vitro* study of specific brain cell populations place particular emphases on gene regulation and the role of growth factors and cytokines in brain development, injury, and repair.

Several investigators at the UCLA MRRC focus on human inborn errors of metabolism and molecular genetics associated with mental retardation and developmental disabilities. Studies of glycerol kinase expression and its binding to porin have helped to elucidate the role of various genes in a particular region of the X chromosome during adrenal development and in mental retardation. Further studies of arginase I and arginase II have the long-term goal of treating diseases due to deficiency of these enzymes by induction or gene transfer techniques.

Using morphologic, metabolic, and behavioral methods, UCLA researchers are studying developmental animal models of both recovery and sparing of function following unilateral brain damage. These studies helped to provide the experimental basis for the clinical treatment of intractable pediatric epilepsy (hemispherectomy), but are also relevant to extensive stroke and cerebral palsy treatments. Other investigators study the behavioral sequelae of traumatic brain injury. These researchers interested in gamma-aminobutyric acid (GABA) and other receptors that predispose people to epilepsy and mental retardation are using a rat model to study epileptogenesis and a mutant mouse model to study Angelman syndrome.

The UCLA MRRC remains one of the major centers for the study of the basal ganglia and its development as well as critical neurotransmitter interactions in development and disease. Researchers are examining the regulation and molecular analysis of the development of brain regions that are sensitive to sex hormones. This group also investigates the role of sex hormones in susceptibility to diseases.

Studies of several behavioral and bio-behavioral investigators at the UCLA MRRC provide insights into the genetics of autism, schizophrenia, and attention deficit disorders. Members of this research group have conducted pioneering studies in behavioral features and intellectual strengths and weaknesses of specific gene disorders. The UCLA MRRC remains a leading facility for the study of children who are at risk for mental retardation and developmental delay and their families, in culturally different ethnic minority populations, including immigrant groups. Members of the socio-behavioral study group have a 30-year history of research on the adaptation of individuals with mild mental retardation and developmental disabilities and their families, studying the developmental

transitions in the lives of mentally retarded and/or developmentally disabled and at-risk children and their families. These investigators have identified the importance of ethnic identity in the successful transition into adolescence. Success in this transition varies widely due to cultural and family origins, particularly among African American youths.

In groups of Latino immigrant youth, behavioral investigators at the UCLA MRRC found that those with greater emergent Spanish literacy development and oral English proficiency at kindergarten are better able to maintain grade level performance in Spanish reading. Socio-emotional development, not cognitive development, appears to affect family adaptation in this diverse ethnic group. Investigators have also found that parents' aspirations for their children adjust to their children's ability levels over time. Successful families focus on family adaptations that promote a better family routine and place less importance on individual stimulation of the child. Whether or not these adaptations change as the children enter adolescence is the focus of current studies being conducted by this group.

THE MRRC AT THE UNIVERSITY OF WASHINGTON (UWA), SEATTLE, WA

Investigators at this MRRC have long had an interest in genetic and environmental causes of mental retardation and developmental disabilities. For almost three decades investigators studying fetal alcohol syndrome (FAS) and related disorders have maintained the most comprehensive and long-term developmental follow-up in the country of people identified as FAS who have been fully characterized. These investigations have systematically uncovered important biomedical and behavioral dysfunctions, which, in turn, stimulated important state and federal activities in intervention and prevention of FAS. For the past 20 years, other investigators at the UWA MRRC have focused their attention on the effects of fetal exposure to methylmercury, using nonhuman primates as models. These investigators have also clarified the association between *in utero* methylmercury exposure and female reproductive dysfunction. The group identified neuropathological effects, such as disorganized lamination and the presence of ectopic cells in the white matter *in vivo* and devised unique experimental strategies to reveal the nature of early cognitive deficits that arise from exposure to methylmercury *in vitro*.

UWA MRRC investigators are mapping and/or isolating genes for the following neurological conditions: Charcot-Marie-Tooth Neuropathy Type 1B, familial Alzheimer disease (Presenilin [PS]1 and PS2 types), and Familial Frontotemporal Dementia.

Investigators at the UWA MRRC have made research advances in the study of epilepsy, at both the single-neuron and simple-circuit levels. This research has identified specific electrophysiological and morphological features that may potentially contribute to

seizure propensity, including the nature of GABA-mediated inhibition in the immature brain, the control of intracellular/extracellular ionic balance, and the vulnerability of immature neurons to seizure-related damage.

Other UWA investigators are examining an immunological basis for the increased susceptibility to infection as well as limitations in responses of the neonate and infant to vaccines. The finding that cytokines, particularly interferon γ, are unduly susceptible has led to new strategies for enhancing antigen presentation and improving and accelerating the immune responses in the neonate and infant.

Behavioral scientists at the UWA MRRC have enhanced the ability to recognize autism in first two years of life. The new neuropsychological and brain-imaging findings in autism indicate that the severity observed reflects different underlying neurobiological bases that can be readily identified; these findings may now help focus early intervention programs. Other investigators in this field have identified and characterized the unique peer interaction deficits experienced by a vast majority of young children with developmental disabilities. Researchers who study early intervention developed a methodology to evaluate parent/child interactions using feeding and teaching scales, a methodology that has been extremely useful in identifying problem areas for children who are at risk. Other studies have successfully enhanced the development of premature, low birth weight children through systematic behavioral interventions during the first three years of life.

UWA scientists are also examining the importance of imitative learning as a foundation for future development in typically developing children, as well as in children with a variety of developmental disabilities. Investigators at the UWA MRRC have identified unique differences between children with Down syndrome and autism, examining possible neurobiological bases for these diseases. New imitative learning procedures that foster development emerged from these studies.

THE MRRC AT THE UNIVERSITY OF NORTH CAROLINA (UNC), CHAPEL HILL, NC

Reorganization of the UNC MRRC enlarged the number of investigators who are involved in MRDD Branch-related research. Many of them study basic mechanisms of central and peripheral nervous system development as well as clinical features of neurogenetic developmental disorders. Using transgenic models and cell culture systems, researchers demonstrated that insulin-like growth factor-1 (IGF-1) promotes brain growth by stimulating proliferation of oligodendrocytes and by reducing apoptosis in neurons. Other work focuses on the critical role that dietary choline plays in the development of hippocampal neurons and memory processes. Researchers are also studying the roles of

neurotransmitters, such as serotonin and GABA, as growth regulatory signals in the developing nervous system, and growth factors, such as nerve growth factor (NGF) and glial derived neurotrophic factor, in the sensory neuron development in the peripheral nervous system. UNC researchers are investigating the role of retinoids in mediating the inductions of the mammalian forebrain and interferon γ, as models for understanding potential pathogenetic mechanisms in neurodevelopmental disorders.

Investigators interested in cell migration and the development of neural connectivity are studying the intracellular signaling cascades that modulate the effects of critical adhesion molecules (e.g., neural cell adhesion molecules, integrins, L1, and cadherins) on the development of neuronal processes. Other researchers are investigating the role of reelin, the product of the *reeler* gene, in inhibiting migration of cortical neurons and enabling detachment of neurons from radial glia, thus effecting normal lamination of the cerebral cortex.

Investigators interested in glial cells and myelination are examining models of abnormal myelination using a mouse model of ceramide galactotransferase deficiency. Other researchers are studying the effects of dietary cuprizone on myelin regeneration.

Long-time members of the UNC MRRC continue their studies of the chemistry, metabolism and function of complex glycosphingolipids, elucidating the role of these compounds in lysosomal storage diseases.

Clinical researchers are using magnetic resonance imaging (MRI) to conduct fundamental studies into the neuroanatomical abnormalities in autism, and were among the first to demonstrate an associated brain enlargement. These investigators were also among the first to identify the broad autism phenotype and its potential importance in genetic linkage studies. This group was also at the forefront of genetic studies in autism, conducting the first large-scale, genome-wide linkage studies. In addition, these UNC researchers conducted the first prospective, longitudinal studies of early development in males with Fragile X syndrome (FXS), through which they identified important temporally defined aspects of the FXS behavioral phenotype.

THE MRRC AT THE JOHN F. KENNEDY CENTER, VANDERBILT UNIVERSITY, NASHVILLE, TN

Investigators at this MRRC are poised on the threshold of a new era in preventing and solving problems in human development. The MRRC organizes its research into four broad areas of biomedical and behavioral research: 1) sensory sciences and neural plasticity; 2) genetics, brain, and behavioral development; 3) communication, cognitive, and emotional development; and 4) learning accommodations for individuals with special needs.

Sensory Sciences and Neural Plasticity

Several investigators at the Vanderbilt University MRRC have made strides in sensory sciences and neural plasticity during the past five years. Their research found that although cellular patterns of brain information processing for adults are established during the embryonic or early postnatal period, environmental enrichment accelerates neuroplasticity, not only in normal animals, but also in animals with stroke-like lesions of the sensory cortex. Furthermore, early sensory deprivation alters somatosensory information processing after maturation. Such effects impact upon the structure and functional organization of the visual and somatosensory systems, an organization that can be influenced by plasticity in cortical and subcortical structures, such as the hippocampus.

Genetics and Brain and Behavioral Development

A number of investigators at this MRRC focus on studying autism and related developmental disorders, such as tuberous sclerosis, Prader-Willi Syndrome (PWS), FAS, and alcohol-related neurodevelopmental syndrome. Some investigators have found that altered serotonin signal transduction is associated with serotonin levels and autistic features. Investigators studying Chromosome 15 have identified a region that is associated with autism. Other MRRC researchers studying PWS have found that children with replication (uniparental disomy) of the PWS gene have an unusual faculty for visual memory that even exceeds that seen in typically developing children. Researchers also found that the obesity observed in PWS children is tied to an interaction between the compulsive eating behavior and a failure of the appetite suppression system. While developing a genetic animal model of autism, another group of MRRC researchers discovered the gene for tuberous sclerosis. Furthermore, other researchers found that exposure to alcohol *in utero* alters the brain's exquisitely sensitive alcohol receptors, results that have direct implications for studies of FAS.

Communication, Cognitive, and Emotional Development

Investigators at the Vanderbilt University MRRC have found new ways to identify children with language disorders, which allows them to refine a differential diagnosis model of autism and subtypes of language disorder. These researchers also devised more accurate measures of prelinguistic skills in children with developmental disabilities as

well as more effective methods for treating speech disabilities in autism and language disorders. These advances have improved methods for treating speech and grammar errors in children.

Investigators studying maternal depression have found that the disorder disrupts the interaction patterns of mothers and their children; such interactions are crucial not only for social and communication development in infants, but also for the continued development of adolescents. MRRC researchers also showed that children living in poverty who have behavioral disorders are more likely to have difficulties with language skills. As a result of these studies, researchers developed a model of stuttering, including linguistic influences on stuttering episodes, that may soon lead to improved treatment for stuttering.

Learning Accommodations for Individuals with Special Needs

MRRC investigators translate discoveries from basic science into direct services for people through a program of learning accommodations for individuals with special needs. Members of this research group have developed innovative methods for teaching mathematics and reading to children with disabilities by including typically developing peers as partners in the learning process. Additional studies of the genetics of intelligence allowed researchers to develop a model for both precocious and general intelligence. Efforts to develop methodology for reducing problem behavior in children and adolescents who exhibit disruptive behavior have aided other investigators in their work to maximize the effective transition for people with developmental disabilities from school, to work, and to daily living environments.

THE MRRC AT THE WAISMAN CENTER, UNIVERSITY OF WISCONSIN, MADISON, WI

Investigators at the Waisman MRRC have made important strides in gene therapy and gene therapy technology in the past five years. Through the development of intra-arterial injection of naked plasmid DNA into muscle and liver, this group has accomplished high levels of gene transfer. Not only does this process mean the potential for foreign gene introduction is now possible in hepatocytes, but such transfer may also prove very useful in the treatment of liver-based disease of inborn errors of metabolism (e.g., phenylketonuria [PKU]). Further, derivation of stem cells from human fetal blastocysts has led to the development of novel methods for growing and genetically manipulating human neural stem cells. Such stem cells, when transplanted into the brain, have the capacity to form new and viable tissue.

Alexander disease is a rare and often fatal disorder of the central nervous system that

usually occurs in young children. Studies of mice that overexpress the gene encoding glial fibrillary acidic protein (GFAP) led investigators at the Waisman MRRC to analyze genomic DNA from patients with Alexander disease and their parents. The investigators discovered nonconservative heterozygous mutations in the coding region of the patients' DNA, but normal DNA in the parents, indicating that mutations in the patients occurred *de novo*.

Disorders of attention in children are often profoundly disabling. During the past year, behavioral scientists at the Waisman MRRC have developed precise quantitative measures of auditory attentional skills that reveal both preschool and school-aged children up to 12 years old have dramatically reduced auditory attention when compared with adults. In addition, researchers can quantify individual differences in visual attention over the first six months of life using the selectivity of visual orientation to a salient object in the visual environment. Further, infants who are near the end of their first year of life are sensitive to conditional probability structures of speech, an ability which provides infants with the means to segment a stream of speech into words.

Investigators at the Waisman MRRC also study strategies by which families care for and support children and adults with developmental disabilities. These families develop resilient patterns of coping during their lifelong caregiving role, patterns that allow them to maintain normative levels of health and psychological well-being without elevated levels of divorce or social isolation. Studies of autism and other developmental disabilities indicate that the behavioral symptoms of autism abate more in adulthood when compared with childhood and the adolescent years. Further, adult individuals with Down syndrome exhibit a pattern of stability in functional skills, behavioral problems, and health instead of a decline with bi-directional influences of parental and child well-being across the life course.

THE MRRC AT THE EUNICE KENNEDY SHRIVER (EKS) CENTER, WALTHAM, MA

Investigators at the EKS MRRC focus on early neuronal migration and differentiation, as well as behavioral studies of individuals with mild-to-profound mental retardation and developmental disabilities. Members of the group at EKS MRRC have a long-standing research interest in glycosylation events that occur during normal and abnormal nervous system development. Glycoconjugates are critical components in normal brain development and play important roles in glycosylation pathways, such as the O-fucosylation of *Notch* receptors. Changes in glycosylation that may involve expression of basic helix-loop-helix transcription factors, like MASH1 and neuroD2, and members of the *Fringe* family of signaling molecules usually accompany the stimulation of stem cell neuronal differentiation.

Several groups at the EKS MRRC are involved in studies of the mechanisms of cell migration in various parts of the developing nervous system and the influence of sex hormones on development. One group is focusing on developing a nonspatial olfactory discrimination procedure to evaluate genetic and sex-related cognitive differences in various strains of mice. In the past, it seemed that prenatal exposure to estrogenic compounds had no apparent effect on olfactory discrimination in male mice; however, EKS MRRC researchers found that perinatal exposure was associated with differential effects, particularly in terms of better-learned olfactory discrimination after reversal learning in mice that were treated with diethylstilbestrol (DES). Effects of both the time course and dosage of DES on adult cognitive ability may reflect alterations that occur during the morphological maturation of the hippocampal granule cells during the perinatal period.

Other investigators at the EKS MRRC are focusing on genetic and epigenetic factors involved in the differentiation of the sexually dimorphic preoptic area/anterior hypothalamus. Although cell migration may begin independent of hormone exposure, GABA and gonadal steroids influence patterns of migration directly when researchers visualized cells in *in vitro* slice preparations by labeling individual cells with the fluorescent dye, DiI. This method enables investigators to study age-dependent changes in cell position and pattern-forming steps that are involved in the development of this area of the brain. Researchers are using similar methodology to study both rostral and caudal cell migrations that occur during distinct stages of olfactory bulb development.

Some investigators at this MRRC are using digital deconvolution microscopy in *in vitro* slice preparations of cells labeled with DiI to study migrating cortical neurons in three dimensions. These investigators are examining the migration of cells from the ganglionic eminence into the cortex, a migration that begins tangential to radial glial guides, and then undergoes a dimensional translation, so that the cells enter the cortical plate radially.

Another group of MRRC investigators has observed highly intricate expression patterns for different retinoid acid (RA) generating aldehyde dehydrogenases in the developing and mature eye. Dynamic expression patterns of retinoid metabolizing enzymes in the eye anlage establish the dorsoventral axis of the retina. Perturbation of RA levels leads to changes in aldehyde dehydrogenase function, while the distribution of the aldehyde dehydrogenases and their level of expression may indicate their role as detoxifying enzymes that protect the eye from light damage. Disturbances in RA levels may alter the increase in RA levels that light produces in the functioning eye and may explain why the developing eye is vulnerable to disturbances in vitamin A.

The EKS MRRC is also concentrating on cerebellar pathology that arises after prenatal exposure to ethanol in an animal model of FAS. Ethanol exposure leads to increased

levels of cerebellar RA *in vivo*. Both ethanol and exogenous RA decrease the production of fucosylated stage-specific embryonic antigen 1, one of a number of glycoconjugates critical for cell migration, adhesion, and survival of cells.

Behavioral researchers at the EKS MRRC focus on the development of cognitive skills in children with severe mental retardation. This research has led to the development of a comprehensive, computer-based methodology for teaching rudimentary reading, spelling, and other symbolic communication skills. These studies demonstrate that educators may reduce or eliminate apparent deficits in learning and perception through carefully designed intervention strategies. One group is studying stimulus control acquisition in individuals with severe or moderate intellectual deficits. These investigators seek to develop a sound theoretical model of transfer and determine the role of attending in effecting transfer of control, both within and across stimulus dimensions. Through these efforts, the investigators hope to develop reliable procedures to teach individuals with mental retardation to relate physically to dissimilar stimuli.

MRRC researchers at the EKS are beginning the first systematic study of social affective functioning in children with PWS, a condition caused by deletion of genes on the q11-q13 region of Chromosome 15. Parents report that children with PWS have difficulty establishing and maintaining friendships with peers. Their behavior is often inappropriate, particularly in response to emotional situations. One potential source of difficulty could lie in their inability to understand and respond to emotional behaviors, as well as their affective responses to another's emotional behavior. Thus, these children appear to have a defect in affective, rather than cognitive, aspects of social functioning.

THE MRRC AT THE UNIVERSITY OF KANSAS (UKS), LAWRENCE, KS

The UKS MRRC has played a major role in generating effective behavioral interventions aimed at the causes, prevention, and treatment of mental retardation, as well as delineating basic knowledge of the underlying biology of typical and atypical development for more than three decades. The Center's research program is organized around four thematic areas that reflect the scientific directions and strengths of the diverse group of involved investigators: 1) language, communication disorders, and cognition; 2) risk, intervention, and prevention of mental retardation; 3) neurobiology of mental retardation; and 4) cellular and molecular biology of early development.

Language, Communication Disorders, and Cognition

Behavioral scientists who study language development in typically developing and developmentally disabled children at the UKS MRRC are focusing on improving language comprehension and production in young, presymbolic children with mental retardation and mediating the effectiveness of prelinguistic communication intervention

through enhanced maternal responsivity. Other researchers have found that, unlike children with Williams syndrome, children with autism show a tense-making deficit in language production similar to children with SLI. Another group of UKS researchers identified kindergarten children who are at risk for later reading difficulties using language factors beyond phonologic awareness that can lead to reading difficulties. Self-instruction among children with mental retardation and limited language skills seems to help mediate generalized problem solving.

Risk, Intervention, and Prevention of Mental Retardation

Center investigators are also studying child and caregiver interactions by identifying components of attention in infants and patterns of mother-infant interaction that are significant in predicting intelligence and cognitive status in later childhood. Because other investigators at the UKS MRRC have found a divergence of strengths and weaknesses of neurocognitive development in studies of ERP waveforms in infants with Down syndrome and FXS, researchers believe that children with developmental disabilities may rely on different components and cognitive strategies. High-quality, non-maternal childcare enhances school readiness skills in children, while young children who are exposed to substance abuse and violence in their caregiving environments have to develop sources of social and emotional resilience that are not needed by other children.

Neurobiology of Mental Retardation

A number of investigators at the UKS MRRC study self-injurious behavior in both humans and animal model systems. Research shows that perseverative behavior is involved in producing temper tantrums in young children with autism, which may contribute to self-injury in these individuals. Other researchers have developed an effective treatment for people with developmental disabilities who self-injure using integrated augmentative communication training and the opiate antagonist naltrexone. Further studies found that treatment with risperidone substantially decreased aggression and self-injury in these patients. Future studies of self-injurious behavior may benefit from the development of a neurophysiologic assessment protocol for universal newborn screening of orofacial and respiratory control in humans. This effort might also benefit from the development of a functional model of sensorimotor entrainment and trigeminofacial modulation in premature and term human neonates.

Using animal models, investigators at the Center demonstrated that training-induced recovery from brain damage in rodents may decrease susceptibility to self-injurious behavior. This led to the development of new instrumentation for quantifying drug-induced behavior at a finer level. In studies of other training-related neuroplasticity in rat models, investigators found that operant training increased brain weight for rodents with microencephaly resulting from prenatal neurotoxicity. Other UKS investigators have developed a non-human primate model of neuro-AIDS that replicates many of the behavioral, neurophysiological, and pathological characteristics of human neuro-AIDS.

Other investigators at the UKS MRRC study neural degeneration, regeneration, and plasticity. These investigators discovered a gene involved in retrograde neuron degeneration and studied the role that neurotrophin-3 plays in the selective rescue of proprioceptive neurons that affects the extent of proprioceptive innervation in skeletal muscle. Further, researchers at UKS revealed that the cause of motoneuron death in the *wobbler* mouse mutant model of ALS is the over-expression of a G-protein-coupled, protease-activated receptor for thrombin. They also found evidence of auditory neuron regeneration following cochlear damage. Other studies are examining the corticospinal system. One group found that red nucleus reorganization contributes to the recovery of motor function after corticospinal axon damage in juvenile rhesus monkeys. Another group found that a behavioral rehabilitation program that involves performing a motor skill task can prevent loss of cortical hand and digit representation that usually occurs with ischemic lesions of primary motor cortex in the monkey. Researchers are also using substrate reduction therapy to successfully slow the disease course in mice with globoid cell leukodystrophy.

Cellular and Molecular Biology of Early Development

A number of investigators at the UKS MRRC are interested in the cellular and molecular biology of early development; to that end, they focus studies on the development of embryo-uterine interactions, placental development and homeostasis, and gene modulation during early development. Thus far, one group identified a zinc-sensing transcription factor that regulates genes involved in zinc homeostasis. Another group determined the critical role of the cyclo-oxygenase-2-derived prostaglandins in embryo-uterine interactions during implantation, as well as signal transduction pathways controlling placental development, and pregnancy specific-modulators of maternal inflammatory cells. Studies of the human placental barrier have not only characterized a functional, multi-drug resistance transporter, but have also established an *in vitro* culture system to investigate drug transport.

THE MRRC AT THE UNIVERSITY OF COLORADO (UCO), DENVER, CO

The UCO MRRC has a unique group of investigators who have studied organic acidemias and lysosomal defects for the past 25 years. Using biochemical, molecular, and genetic studies in relevant animal models, these investigators have identified genetic defects and pathophysiological mechanisms in glutaric acidemia (Type I), glutaric aciduria Type II, D-2-hydroxyglutaric acidemia, homocystinuria, and propionic aciduria, and have developed strategies for treating these disorders. Specific organic acid screening assays developed by investigators at the UCO MRRC are used worldwide for diagnosing these disorders in children.

Other investigators at the UCO MRRC are conducting basic studies of synapse formation and function. Investigators studying molecular aspects of nerve growth cone development found that a novel IGF-1-receptor β-subunit serves as a ligand to initiate an intracellular signaling cascade that controls plasmalemmal expansion and helps to regulate pseudopod motility and attachment. Another group of researchers, studying the effects of mutation on one particular voltage-gated sodium channel, sodium channel 6, has found that this channel is mutated in both the *med* (motor endplate disease) and *jolting* (cerebellar ataxia with Purkinje cell loss) mutations, mutations that produce very different and distinctive movement disorders.

A number of investigators at the Center study genetic and behavioral aspects of Down syndrome using the partially trisomic Ts65Dn mouse model. They have found that these mice display deficits in learning and memory similar to those of individuals with Down syndrome, and suggestive of deficits in the prefrontal cortex and hippocampus. Although resting membrane potentials in the hippocampus increased significantly in these models, the investigators found no deficits in long-term potentiation. Unexpectedly, investigators did find that Ts65Dn mice were superior to normal mice on a rotorod locomotion task that is designed to train animals gradually, but were not when the task does not involve gradual shaping. These researchers also found that cholinergic deficits that appear in these mice by six months of age arise from a lack of cholinergic marker expression by basal forebrain neurons, not the premature neuronal death. Currently the group is using a neurotrophin that is normally produced by the hippocampus, NGF, coupled to the transferrin receptor for treating these cholinergic deficits directly.

The UCO MRRC has focused a great deal of attention on particular genes located on human Chromosome 21, among them *RED1* and *GART*. Mice transgenic for the *RED1* gene, which plays a role in editing glutamate and serotonin receptors, exhibit not only different levels of editing, but also region-specific differences in alternative processing of the gene. In addition, investigators interested in purine metabolism have created several useful animal models. For instance, they are comparing purine metabolism in mice transgenic for the purine biosynthetic enzyme cascade *GART* with uricase-deficient mice and mice with inactivated reduced folate carrier (REFC). While the investigators found that complete inactivation of *REFC* is a lethal condition in mouse embryos, eliminating the expression of the Fragile X mental retardation protein (FMRP), encoded by the gene *FMR-1*, is not lethal. Mice homozygous for mutation of *FMR-1* exhibit an accentuated auditory startle, display startle at lower amplitudes of sound, and exhibit accentuated pre-pulse inhibition at lower prepulse levels. Although less hyperactive than Ts65Dn mice, *FMR-1*-deficient mice have greater ability to inhibit activity in open arms of a plus maze, an ability also found in normal mice.

Behavioral scientists at the UCO MRRC focus on several conditions associated with mental retardation and developmental disabilities: autism, FXS, schizophrenia, and Down syndrome. Investigators who are conducting longitudinal studies of the phenotype

of autism at behavioral and neuropsychological levels have found that young children with autism do not show significantly more sensory-related behaviors. Children with autism do, however, show a different pattern of relationships between sensory functioning and functioning in other areas. The investigators found that sensory behaviors in autism are related to both the degree of withdrawal from novel situations and the level of motor functioning. Researchers studying children with FXS who exhibit autistic symptoms have found further evidence for genetic influences.

In studying genetic predisposition to schizophrenia, UCO researchers have found a marker for genetic risk that allows them to identify the disease in children as young as six, long before disease onset. The marker, which is the tendency to anticipate target motion during a smooth pursuit eye movement task, is linked to a high-affinity nicotinic acid receptor on Chromosome 15p. Further, evidence from studying the types of comorbid psychopathology that occurs in children of schizophrenics reveals Attention Deficit Hyperactivity Disorder (ADHD), anxiety disorder, unipolar depression, and primary psychotic disorder, usually schizophrenia, and particularly, childhood-onset schizophrenia.

Several groups of investigators study phenotype-genotype relations in reading disabilities and dyslexia. Studies indicate that although reading disability (RD) and phonological disorder (PD) differ in their age of onset, they overlap phenotypically. Prospectively, children with PD later have elevated rates of RD; retrospectively, children with RD are likely to have had elevated rates of PD. Further, the underlying phonological processing disorder that is characteristic of RD also occurs in many children with PD. Investigators at the UCO MRRC who had previously identified a dyslexia-related gene on Chromosome 6 have expanded their study to include other genes, such as some associated with ADHD, the dopamine transporter 1 (DAT1), and the dopamine receptor D4 (DRD4). Investigators are currently examining differences in state-of-the-art component reading skills in their genome-wide search for dyslexia loci. These investigators also found individual differences in the sizes of specific brain structures and brain correlates of RD children, when they compared structural MRIs of twins with RD with MRIs from controls.

The UCO MRRC has begun developing neuropsychological profiles of individuals with Down syndrome. When investigators analyzed prefrontal cortex, hippocampus, and cerebellum, they found selective deficits on hippocampal, but not on prefrontal measures, in Down syndrome, a pattern similar to that of the Ts65Dn mouse. Additional studies examine the neuropsychological phenotype associated with autism, by looking at the extended phenotype of the parent and using magnetoencephalography (MEG) to study brain function in adults with autism. Currently, these investigators have a particular interest in longitudinal study of autism in individuals with FXS and Down syndrome.

THE MRRC AT THE BAYLOR COLLEGE OF MEDICINE, HOUSTON, TX

For a number of years, investigators at the Baylor MRRC have mapped and identified genes that cause severe mental retardation and developmental disabilities: X-linked incontinentia pigmenti (IP); monosomy 1p36; Angelman syndrome; type-1-spinocerebellar ataxia (SCA1); Rett syndrome; PWS; FXS; Smith-Magenis syndrome; velo-cardio-facial syndrome (VCFS); Friedreich's Ataxia; two X-linked immunodeficiencies; and X-linked chondrodysplasia.

Baylor MRRC studies show 85 percent of X-linked dominant IP cases involved a particular mutation in the gene *NEMO*. Lethal to males *in utero*, mutations of *NEMO* in affected girls lead to disparate symptoms, such as widespread skin blistering and pigment problems, dental abnormalities, hair loss, mental retardation, and vision problems due to retinal hypervascularization. Different types of mutations in the *NEMO* gene may manifest themselves as distinct clinical disorders, including immunodeficiency. The *NEMO* product helps to regulate the activities of NF-kB, a key molecule in a signal transduction pathway that affects many cellular functions. Without the function of the *NEMO* protein, cells are unresponsive to the myriad signals that are normally communicated through NF-kB.

Investigators at the Baylor MRRC recently recognized a new and, very likely, contiguous gene syndrome caused by monosomy 1p36. Monosomy 1p36 is probably the most common terminal deletion syndrome contributing to mental retardation in our population, with an estimated incidence of one-in-5,000 newborns. The majority of monosomy 1p36 patients have mental retardation, seizures, and sensorineural hearing loss with varying degrees of severity. Specific genes located in the 1p36 chromosomal region also appear to be responsible for the epilepsy seen in this syndrome, as well as the hereditary hearing loss and mental retardation in these patients.

While Angelman syndrome can arise from different genetic mechanisms, one of the most common is a mutation of *UBE3A*, the gene that encodes the enzyme E6-AP ubiquitin ligase, located on Chromosome 15. Patients with Angelman syndrome display an odd constellation of symptoms: mental retardation, seizures, ataxia, hand-flapping, inappropriate laughter, and occasionally, a strange fascination with water. Mutations in the *UBE3A* gene disable an enzyme that normally "tags" proteins fated for recycling within the cell, which leads to the accumulation of probably only a few specific target proteins within the cell. This accumulation is responsible for the clinical features of Angelman syndrome, which investigators are now studying using an animal model developed at Baylor.

Other investigators at Baylor have exploited the Angelman syndrome mouse model in studies that have had a huge impact on research into the pathogenesis of several

neurodegenerative disorders, such as SCA1. When *Ube3a* mutated mice are mated with SCA1 mice, the protein accumulation from impaired protein recycling exacerbates disease symptoms in this adult-onset condition.

Last fall, researchers at the Baylor MRRC described mutations in the X-linked gene encoding methyl CpG-binding protein 2 (MECP2) that were associated with 60 percent to 75 percent of cases of Rett syndrome. Normally, *MECP2* encodes a protein that "orchestrates" gene silencing at different, but precisely regulated, times and places in the body via several different mechanisms. Mutations in *MECP2* render the protein unable to "conduct" gene expression, so some genes express their products at the wrong time, creating developmental chaos. Dysfunction of MECP2 is associated with a wide variety of other disorders, ranging from mild learning disabilities in adult women, to severe neonatal encephalopathy in male infants. Utilizing a previously developed mouse model, investigators at Baylor are now poised to probe which genes are targeted for silencing under this particular mechanism. Further, because the dysfunction affects methylation, therapeutic trials with drugs like folate and betaine are currently underway at the Rett Syndrome Center at Baylor and the University of Alabama at Birmingham. (See the section on the MRRC at the University of Alabama, Birmingham, for more information).

THE MRRC AT THE KENNEDY KRIEGER INSTITUTE, BALTIMORE, MD

Imaging studies done by investigators associated with this MRRC have led to the development of a high-resolution MRI protocol for measuring amygdala and hippocampus that may apply to studies of individuals with various types of mental retardation and developmental disabilities. In FXS patients, for example, investigators have found enlargement of the hippocampus. In addition, they also used a semi-automated parcellation method (SAPM) in studies with children with several mental retardation and developmental disabilities-related syndromes. Patients with FXS, for example, exhibit differences in cerebral cortical growth curves between full mutation and mosaicism. Patients with VCFS have posterior cortical white matter involvement. Their magnetic resonance spectroscopy (MRS) studies also reveal white matter involvement in patients with Rett syndrome. Although these investigators were among the first to study macrocephaly in neurofibromatosis type 1 (NF-1), their recent MRS studies led them to redefine the neuroimaging abnormalities in NF-1 in terms of the basis for thalamic involvement. In addition, researchers are using the relatively new and sensitive method of diffusion tensor imaging to delineate and reconstruct fiber pathways in normal adults and, now increasingly, in studies of individuals with mental retardation and developmental disabilities or related conditions.

Continuing their long history of studies of inborn errors of metabolism, investigators at this MRRC have identified a defective form of the gene that encodes α aminoadipic semialdehyde located on the q31.3 region of Chromosome 7. Patients with this mutation have familial hyperlysinemia, an autosomal recessive disorder associated with mental retardation, hypotonia, seizures, and impaired sexual development. Further studies show that a predisposition of genomic structure to rearrangement in the regions flanking the gene *ORNT*, encoding the mitochondrial ornithine transporter, leads to microdeletions, which in turn give rise to hyperornithinemia-hyperammonemia-homocitrullinemia (HHH) syndrome. Finally, inducing genetic abnormalities in the gene that encodes the peroxisomal membrane protein, PMP70, also induces massive medium-chain dicarboxylic aciduria in the plasma and urine of mice.

THE MRRC AT THE UNIVERSITY OF PENNSYLVANIA (UPA), PHILADELPHIA, PA

UPA MRRC studies of ornithine transcarbamylase (OTC), a urea cycle enzyme whose deficiency causes mental retardation, have continued since the last MRDD Branch report to NACHHD Council. Specifically, these researchers replaced the mutant OTC gene with an adenoviral vector that contained a normal copy of the gene. Although approved to conduct gene therapy trials in 1994, their gene therapy studies were recently suspended, due to the untimely death of an OTC patient participating in the clinical trial.

New investigators recruited to the UPA MRRC are involved in studies of early patterning events in the nervous system, as well as studies of the molecular basis of the transition from short-term to long-term memory and of the molecular basis of increased excitotoxic neuron damage in the presence of calcium-activated proteases. Several MRRC investigators are also involved with studies of gene effects on early patterning events in the nervous system, with emphasis on the genes *Sonic hedgehog (Shh)* and *bone morphogenetic proteins (Bmps)*. Expression of *Shh* and other genes are important in the growth and differentiation of the ventral neural tube, processes that are modulated by the expression of genes like the *Bmps* in the dorsal neural tube. Overexpression of *Bmps* causes increased cell death in the ventral neural tube midline, which leads to a condition reminiscent of holoprosencephaly. Investigators studying the role of signaling pathways that involve protein kinase A (PKA) in converting short-term memory to long-term memory storage are now able to visualize and correlate expression with neural activity in real time. They can then qualify these measurements using positron emission tomography (PET), fMRI, and autoradiography. Researchers are also studying the calcium-activated protease, calpain, which acts on the glutamatergic NMDA receptor to increase the amount of neurotoxic damage observed. Rather than changing the ion channel properties of the NMDA receptor, calpain dysregulates the activity of the receptor by releasing its subunits, particularly the NR2B subunit. These subunits have increased mobility in the cell membrane and thus expand the area of excitotoxic insult.

THE MRRC AT THE UNIVERSITY OF CALIFORNIA (UCI), IRVINE, CA

Investigators at the UCI MRRC have demonstrated an association between a dopamine D4 allele and ADHD, with more severely affected children showing a particularly strong association. MRI studies at the Center show significant differences between the brains of children with Down syndrome, autism, or ADHD and the brains of controls. Specifically, changes in brain size and/or white matter constitute the first real evidence of pathophysiology in these conditions. These findings provide the basis for developing hypotheses for future research. Further, applying new tools, such as MRI and fMRI, will allow these investigators to continue to develop insights into areas not previously explored.

THE MRRC AT THE UNIVERSITY OF ALABAMA (UAB), BIRMINGHAM, AL

Investigators at the UAB MRRC are involved in a variety of basic, clinical, and behavioral studies relevant to mental retardation and developmental disability. Some study processes that mediate memory and learning, such as synaptogenesis and synaptic plasticity in the mature nervous system, while others study rare and sometimes devastating neurological diseases, like Alexander disease. Still others are conducting clinical trials for Rett syndrome and glioblastoma multiforme. Behavioral scientists at the UAB MRRC focus their studies on adolescents with mild mental retardation.

Several researchers at the UAB MRRC are investigating processes that mediate memory and learning, particularly in the cerebral cortex and hippocampal formation. Some of these studies focus on molecules involved in axon guidance and synapse formation, while others examine the roles of neurotrophic factors, such as brain-derived neurotrophic factor (BDNF) and neuroregulin, in both normal and abnormal brain development. Center researchers discovered that a release of nitric oxide from a rich plexus of axons maintains patterns of connectivity and corrects mistakes that may occur. This plexus of axons arises from a population of neurons that are located in the white matter underlying the cerebral cortex. These neurons survive the period of intensive cell death that occurs during late fetal and early postnatal development. Thus, these neurons can facilitate learning and memory in both the immature and the mature brain. Disruption of these neurons is associated with a number of neurological diseases, including some forms of mental retardation.

UAB researchers are studying another molecule that is important in initiating synaptogenesis. They are exploring the function of the product of the gene *Bassoon*, a protein whose precursor is expressed at the presynaptic active zone prior to the formation of the postsynaptic density. Once formed, docking of synaptic vesicles at the active zone is facilitated by BDNF, through the tyrosine kinase receptor, TrkB. By promoting

formation for new synaptic contacts, BDNF helps to increase dendritic spine density on individual hippocampal pyramidal neurons, an effect that may prove useful in triggering the growth of new brain circuits after injury or deprivation during development. Neuroregulin acts through ErbB protein tyrosine kinase receptors located on the postsynaptic side of the synapse. In addition to its key role in development, persistence of neuroregulin signaling in the mature nervous system may help to regulate synaptic efficiency during learning. Lack of either neuroregulin or its receptor leads to profound malformation of brain and early death.

Other UAB investigators, like their colleagues at the Waisman MRRC, are focusing on Alexander disease. As mentioned earlier, Alexander disease is a devastating, often fatal disease of infancy caused by *de novo* gain of function mutations in the gene that encodes GFAP. Thus, Alexander disease is the first disease identified as a primary disorder of astrocytes, the first disease shown to be due to a mutation of *GFAP*, and the first disease reported to involve a "gain of function" mutation of a gene encoding an intermediate filament protein.

Other members of the UAB group continue their pioneering studies of Rett syndrome, in parallel with the MRRC group at the Baylor College of Medicine. These researchers have recently initiated a blinded, placebo-controlled clinical trial using folate/betaine to modulate methylation in patients with Rett syndrome, particularly those with mutations in the *MECP2* gene. Other important clinical trials underway at UAB are testing the biosafety of a naturally occurring toxin which has particularly high affinity for malignant glial cells in the primary brain tumor, glioblastoma multiforme.

CHROMOSOME ABNORMALITIES

Another important facet of MRDD Branch research focuses on chromosomal abnormalities and their resulting effects and conditions. Chromosome abnormalities account for a substantial amount of morbidity and mortality in children. Approximately 20 percent of mental retardation is attributable to cytogenetic abnormalities, which can include trisomy, mosaicism, or chromosome rearrangements, such as translocations, terminal deletions, interstitial deletions, or interstitial duplications. Recent advances in high-resolution cytogenetic analysis and the development of the molecular cytogenetics field, mainly through the application of fluorescence *in situ* hybridization, or FISH, provide opportunities for identifying subtle chromosomal rearrangements, such as cryptic deletions, as the causes of a number of genetic syndromes. Using these modern technologies, Branch-supported researchers have shown that some regions of the human genome seem to be more prone to rearrangement than others. The following information describes MRDD Branch research on chromosomal abnormalities.

WILLIAMS SYNDROME (WMS)

WMS is a genetic disorder with an approximate prevalence between one-in-10,000 and one-in-20,000 people. Studies of WMS offer the unique opportunity to understand links among genetics, cognitive abilities, and underlying brain development. WMS arises from the deletion of a contiguous group of genes on the long arm of Chromosome 7, including the *elastin* locus in 7q11. Cardiac, sensory, and cognitive defects, as well as distinctive facial features, abnormal calcium metabolism, failure to thrive during infancy, and moderate levels of mental retardation characterize many individuals with WMS. Molecular advances make it possible to establish a diagnosis in 95 percent of cases.

Selective sparing of abilities, such as the ability to recognize, discriminate, and remember familiar and unfamiliar faces, accompanies spatial defects in WMS, making it a good model for determining the minimal cognitive substrates needed for developing language and social cognition. Patients with WMS have relatively good communicative skills and are usually gregarious; further, they are highly sociable and tend to be overly familiar with strangers, a characteristic similar to that found in individuals with frontal lesions. In some respects, persons with WMS may be the cognitive, social, and neural opposites of individuals with autism, although the majority of adults in both groups are unemployed and usually require supported environments.

The MRDD Branch supports studies in WMS that examine the links among the domains of genetics, cognition, language, affect, memory, attention, spatial, social, and musical ability. Branch researchers use a variety of techniques to study brain and cognitive relations, including ERPs during language and face processing and MRI using quantitative 3-D analysis. Some investigators report that although face processing and auditory language comprehension are relatively spared in WMS, they observed specific ERP patterns in WMS patients that are not characteristic of normal development at any age or of any other clinical population.

Researchers correlate these studies with anatomic studies that reveal characteristic differences between WMS and Down syndrome brains. In WMS, limbic and neocerebellar regions are relatively spared when compared with Down syndrome. Interestingly, the regions that are spared in WMS are dysplastic in patients with autism. Posterior regions, particularly those involved in visuo-spatial functions, also appear to be curtailed in WMS patients. Further, WMS patients exhibit increased neuronal size and decreased neuronal packing density, perhaps in association with abnormal neuronal survival and/or abnormal connectivity.

Functional spatial deficits in WMS are similar to those of patients with early focal lesions of the right hemisphere of the brain; the deficits, however, are more severe and persistent in individuals with WMS. Within certain explicit memory tasks, WMS patients'

phonological skills appear to be stronger than their visuo-spatial skills, while verbal working memory is well preserved relative to general intelligence. Both WMS and matched Down syndrome adolescents fail cognitive conservation tasks that are easily mastered by young normally developing children. Adolescents with WMS, however, outperform their Down syndrome peers on grammatically complex sentence forms, comprehension, and production of affect. When WMS patients are tested for implicit learning, such as artificial grammar learning, they perform less well than normally developing individuals, when matched for vocabulary level, but show evidence of learning artificial grammar. Thus, this distinctive cognitive process is partially impaired in WMS, in contrast to phonologic memory processes in the same patients.

DOWN SYNDROME

A landmark accomplishment, the completion (99.7 percent) of the sequence of human Chromosome 21, was announced in May 2000. The anticipated date for converting the draft to final sequence of the approximately 240 genes located on Chromosome 21 is spring 2001. Having the final sequence will enable investigators not only to identify genes relevant to Down syndrome, but also to facilitate creation of animal models with which to study the effects of altered expression of specific genes located on Chromosome 21 on growth and development of various organs, particularly the brain.

In the past several years, researchers have made substantial progress on the development and characterization of such animal models, including the generation of animals with segmental trisomy for only the portion of human Chromosome 21 associated with the phenotypic characteristics of Down syndrome when present in triplicate. Comparisons of animal models, such as the Ts65Dn and Ts1Cje (Ts108Cje) partial trisomic mice, are being conducted in a systematic and concerted manner. Such studies have enabled researchers to focus on small groups of genes that may be associated with specific common characteristics of the syndrome, such as congenital heart disease, cholinergic neuron dysfunction, hippocampal growth factor deficiencies, and cognitive deficits that worsen in Down syndrome individuals with aging. In Ts65Dn mice, for example, studies reveal an acquired cholinergic hypocellularity is associated with learning and memory deficits, deficits that are comparable to those observed in Down syndrome individuals.

Such studies have enabled one group of investigators to initiate research using stem cell therapies on Ts65Dn mice, including the use of MRI and microPET imaging of animals transplanted with cells containing magnetic nanospheres and optical/fluorescent molecules, respectively. Such marker molecules or structures allow non-invasive imaging of the transplanted cells in the live, awake animal in real-time. Although currently limited by the number of Ts65Dn animals available for study, success in these experiments will lead to the next steps: the definition of the brain region(s) critical for

the improvements seen in the grafted animals, and determination whether it is the cells themselves or factors provided by the cells that are responsible for the improvements.

In addition to these animal studies, investigators studying the effects of aging in elderly Down syndrome patients have begun to systematically chronicle alterations in cognitive and motor function that may accompany the inevitable development of the pathological features of Alzheimer disease in this population. Such alterations do not begin in the adult period, but are evident during the first year of life when cognitive, linguistic, and gross motor domains exhibit the fewest and least-consistent gains actualized through early intervention. Deficits in young children with Down syndrome occur in the auditory memory span, intelligibility, and specific language delay, with more severe effects on syntax than semantics, which improve during adolescence. Long-term, home-based training to increase working memory efficiency improves working memory in Down syndrome children. Young adults with Down syndrome exhibit specific verbal-motor difficulties. Persons with Down syndrome learn and retain sequential movement skills better if they are taught with a visual, as opposed to a verbal, instructional protocol.

For children with disabilities, family interactions and the manner in which family members help children cope with social rejection will influence the children's abilities to process social information and solve social problems. Other investigators are seeking to help Down syndrome adolescents collaborate effectively by identifying ways that developments in nonverbal cognition, language, social cognition, speech, and hearing domains contribute to discourse with other adolescents.

X CHROMOSOME DISORDERS

X-Linked Adrenoleukodystrophy (X-ALD)

X-ALD is a serious genetic disorder that affects the white matter of the nervous system, the adrenal cortex, and the testes. The minimum frequency of affected males in the population is one-in-42,000, while the combined frequency in men and women is one-in-18,000. Affected boys develop normally until four-to-eight years, after which they suffer dementia and a progressive neurologic deficit that leads to a vegetative state. The adrenal insufficiency presents as Addison disease, responding readily to steroid replacement. The principal biochemical abnormality is the accumulation of very long chain fatty acids (VLCFA) due to an impaired capacity to degrade them, a reaction that normally occurs in the peroxisome. The defective gene is a peroxisomal membrane protein that maps to the q28 region of the X chromosome. The defect involves an impaired capacity to generate the coenzyme A (CoA) derivative of VLCFA in the peroxisome, a reaction that is catalyzed by very long chain acyl CoA synthetase. Diagnosis is based on demonstrating increased levels of VLCFA in plasma, red blood cells, or cultured fibroblasts. The biochemical abnormality is already present on the day

of birth. Doctors can reliably diagnose women who are heterozygous for X-ALD using a newly developed DNA-based assay. A significant proportion of heterozygous females is symptomatic for the disease. Prenatal diagnosis of affected hemizygotes was first accomplished in 1982, through a demonstration of increased levels of VLCFA in cultured amniocytes and chorionic villi. To date, therapies include: dietary treatment with a mixture of glyceryl trioleate and glyceryl trierucate (referred to as Lorenzo's Oil), immunosuppression, β interferon, and bone marrow transplant (BMT). Although BMT is the only proven effective treatment among these therapies, it is associated with severe complications, including death. Eight-to-10 year follow-up of patients with the childhood cerebral form of X-ALD who received BMT in the early stage of the disease indicates that this treatment often leads to stabilization and, occasionally, to reversal of abnormalities. Recently, doctors proposed using Lovastatin as a therapy because it corrects, at least in part, the defect in VLCFA metabolism in cultured fibroblasts of patients with X-ALD. Studies in the mouse model of X-ALD have shown that 4-phenylbutyrate treatment can also lead to reduced VLCFA levels in the brain and adrenal glands and normalized biochemical abnormalities, providing hints to new forms of therapy.

Rett Syndrome

Rett syndrome is a neurodevelopmental disorder that primarily affects females, but is also a global syndrome: no population or ethnic group is unaffected by this rare, but devastating, disorder. Although development appears normal for the first six-to-18 months of life, an estimated one-in-10,000 to one-in-15,000 girls then begin to lose their speech and purposeful hand movements and to engage in stereotypic hand wringing movements that may be accompanied by hand biting or mouthing. These girls exhibit acquired microcephaly, ataxia and apraxia, seizures, and episodic hyperventilation followed by episodes of apnea or "breath-holding." The symptoms of Rett syndrome reach a plateau several years after the onset and does not progress further during the life span, although it is often associated with sudden and unexpected death in affected girls.

Originally described by Andreas Rett in 1966, little was known about Rett syndrome until exclusion mapping led to the localization of the Rett syndrome gene to the X-chromosome and eventually to the Xq28 region within the last five years. Although the involved gene(s) underwent X-chromosome inactivation, the process of inactivation differs in affected girls and in asymptomatic/obligate carriers, differences that were revealed through studies of methylation.

One gene involved in methylation located in Xq28 is *MECP2*, a gene that encodes a methyl-CpG binding protein 2. Mutated *MECP2* is unable to switch off a group of genes, which encodes otherwise beneficial proteins. When these proteins are present in excess, however, the survival and function of cells in the nervous system are altered profoundly and progressively. As these proteins accumulate, chaos begins, although declines due to the chaos are often not evident before the second year of life. Males, who possess but

one X chromosome, have no defense mechanism against this accumulation of proteins; almost all males with the disorder die during gestation or during infancy, which accounts for the almost exclusively female distribution observed. During the past year, however, numerous comparative studies around the world have verified that only 60 percent to 75 percent of females with Rett syndrome have mutations in *MECP2*. Further, at least one pedigree of severely retarded males with distinctive facial phenotype has survived into adolescence with the same mutation in the *MECP2* gene as that found among some females with Rett syndrome. Thus, it is possible that the *MECP2* gene is only one gene whose mutation causes Rett syndrome, while mutations in the *MECP2* gene are not uniformly fatal to males who possess them. Because *MECP2* is expressed early in development, and mice mutant for *MECP2* fail to complete development to be born live, studies directed toward the manipulation of the *MECP2* gene that would enable slowing, reversal, or even prevention of the progression of Rett syndrome are of great interest. Currently, two clinical trials are underway utilizing drugs like folate and betaine, safe drugs that modulate methylation.

Fragile X Syndrome (FXS)

Over the past 10 years, FXS has emerged as the most common inherited cause of mental retardation. Investigators supported in part by the NICHD have determined that FXS is an X-linked dominant condition with reduced penetrance. FXS occurs in all ethnic groups with a frequency of one-in-1,250 males and one-in-2,500 females. Investigators supported in part by the MRDD Branch found that FXS results from a mutation in a gene located in the q28 region of the X chromosome, now called Fragile X Mental Retardation-1 (*FMR-1*). The mutational change that occurs in nearly all affected patients is the unstable expansion of a cytosine-guanine-guanine (CGG) trinucleotide repeat in the 5' untranslated region of the *FMR-1* gene. The length of the trinucleotide repeat determines the severity of the phenotype observed, ranging from normal (<50 repeats), to premutation, to affected or full mutation (>200 repeats). Although full mutation affects both males and females, premutation and full mutation females may exhibit marked variability in phenotypic characteristics, depending on the extent of inactivation of the affected X chromosome.

Although most children with FXS require a lifetime of special care at immense expense, the FXS phenotype shows considerable variation, ranging from near-normal functioning to severe cognitive impairment, with particular deficits in visual-spatial perception, speech and language, attention, self-regulation, and short-term memory. Recently, investigators have studied the responses of adolescents with FXS to several tasks, such as nonverbal memory and problem solving ability, auditory memory, repetitive language skill, expressive language skill, and perspective-taking ability. The results have helped investigators identify ways in which developments in nonverbal cognition, language, social cognition, speech and hearing domains contribute to the ability of these adolescents to collaborate effectively. The results also allowed researchers to determine whether these domains make different contributions to discourse in Down syndrome and FXS patients. Other investigators have studied the impact of FXS on early

neurocognitive development among infants, toddlers, and preschoolers. By analyzing ERPs recorded from infants and children with FXS, investigators identified syndrome-specific strengths and weaknesses with respect to attention, expectancy, stimulus encoding, sequential information processing, visual recognition memory, and social gaze.

Other studies, supported in part by the Institute, provided important insights into the unique manner in which the trinucleotide repeat mechanism gives rise to mutation of the *FMR-1* gene and alters the function of its protein, FMRP. Investigators have found that the repeat length of the unmethylated and expressed alleles on the active X chromosome increases levels of FMRP. As the length of the messenger RNA (mRNA) transcript that is produced increases, transcriptional efficiency decreases and *FMR-1* gene expression becomes upregulated. Investigators found that although abnormal methylation patterns may be present from the start, the expanded *FMR-1* gene is inactivated by deacetylation at specific histones. Some investigators have used this information to design therapeutic interventions involving 5-azacytidine to increase acetylation and promote gene reactivation. Others are focusing their efforts on studying specific methyltransferases whose activity may be altered during the abnormal methylation process.

Investigators studying the normal function of FMRP have found that the protein exists in both nucleus and cytoplasm and have used several methods to localize FMRP as it shuttles between nucleus and cytoplasm. In the cytoplasm, they found FMRP associated with a polyribosomal ribonucleoprotein (RNP). These polyribosomes exist not only in the cell body, but also in dendritic spines in close association with synapses, which implies that FMRP may play a role in synaptic transmission and plasticity. Investigators found that mutant FMRP sequesters bound mRNAs, preventing their translation or the translation of interacting messages. These researchers also determined that at least six proteins are found in FMRP-containing RNPs, among them, FMR family members FXR1 and FXR2 and the protein nucleolin P2.

Over the last five years, researchers have gained much insight into the origins of the trinucleotide repeats. Investigators determined that these repeats are not embryonic in origin, as was originally expected, but appear to be oocyte-derived and are present in the primordial germline prior to sexual differentiation. In the developing testis, for example, full mutation repeat lengths contract to premutation size at an accelerated manner; while scientists have yet to establish the effect on the developing ovary, mutation repeats may be a factor in the genesis of the markedly premature menopause (<29 years of age) that is experienced by some female premutation carriers. Continuing studies have determined that investigators are more likely to find premutation carriers in a population of females with markedly premature menopause (<29 years of age), rather than among those with early menopause (35-42 years of age). Further, individuals with premature ovarian failure who do reproduce have a higher frequency of children with chromosomal abnormalities, such as Down syndrome.

A group of investigators supported in part by the MRDD Branch created mice with null mutations of mouse *FMR-1* genes, expecting them to exhibit features of the phenotype of FXS individuals. Unexpectedly, these mice exhibited a relatively benign phenotype. Like FXS individuals, however, these mice do exhibit cognitive deficits in short-term memory and visual-spatial tasks. They also exhibit some cellular phenotypes that seem consistent with FXS, such as delayed synaptic remodeling, observed in cultured neurons derived from the hippocampi of these mice. Such observations led investigators to better appreciate that FXS appears to be a human-specific disorder, since the repeat lengths in other mammals are either very short, or highly interrupted. Current efforts to create a model that more faithfully reproduces the features of FXS focus on the generation of mice with fragments of human X chromosomes that contain pre- and full-mutation repeats through irradiation and microcell transfer into embryonic stem cells.

The *Workshop on Fragile X: New Research Directions* held in December 1998, stimulated the release of a request for application (RFA) entitled "Neurobiology and Genetics of Fragile X Syndrome" [HD-00-015] in April 2000. A substantial number of both small, short-term, innovative pilot research grants as well as larger, longer-term, investigator-initiated grants were received in response to the RFA. A special emphasis panel recently determined the scientific merit of these proposals, recommending several for awards that may commit as much as $1.4 million annually in total costs for the next five years by the NICHD, the National Institute of Mental Health (NIMH), and the Fragile X Research Foundation (FRAXA). The NACHHD Council will consider grants with sufficient scientific merit for funding in January 2001.

PRENATAL MALNUTRITION

Malnutrition remains one of the most prevalent conditions affecting children. Periods of mild-to-moderate malnutrition prior to two years of age are associated with delays in cognitive development and poor performance in school later in life. Environmental factors, such as poverty and infection, may occur simultaneously with malnutrition, and confound the results obtained by a majority of studies in human populations. Researchers have consequently depended on animal models to provide detailed information on the effects of malnutrition on the developing brain. Investigators supported by the MRDD Branch are using a multidisciplinary approach to contribute significantly to our understanding the effects of prenatal malnutrition on cognitive development and brain function. The rodent model they employ recreates as closely as possible the conditions that are known to contribute to the occurrence of small-for-date babies in human populations.

To investigate intergenerational effects of malnutrition, investigators use rats whose mothers were malnourished before and/or during pregnancy. Malnourished rats then receive a restricted amount of protein in the five weeks prior to pregnancy and in the three weeks of pregnancy. The test and control diets are isocaloric with identical amounts of vitamins and minerals. At birth, the experimental pups are rehabilitated nutritionally by cross-fostering them to control dams. Pups that result from this model are small in size relative to well-nourished control pups, but are otherwise healthy. Investigators follow the rats over the life span and test them developmentally from embryonic stages to adulthood.

These studies show that prenatal malnutrition adversely affects cognitive performance, emotional reactivity (including stress responsiveness), and social interactions. Prenatal protein malnutrition particularly affects the inhibitory neurotransmitter, GABA. Negative effects are also observed in *in vivo* long-term potentiation, kindling, paired-pulse measures, and *in vitro* miniature inhibitory postsynaptic currents.

In addition, prenatally malnourished rats respond abnormally to a variety of psychological and physiological stressors, which led investigators to examine the effects of stress on measures of neurobiological development and function. In the neonatal malnourished rat, stress produces excessive activation of the immediate early genes that regulate brain function. In prenatally malnourished rats, stress in the neonatal period also alters two important processes in the hippocampal cortex—the generation of neurons by cell division and the pruning of excess neurons by apoptosis. Despite the relative preservation of prenatal neuron development in the brainstem (locus coeruleus and raphe nuclei), postnatal processes that provide the basis for the plasticity of the normal brain are, in fact, disrupted.

MATERNAL PHENYLKETONURIA (MPKU)

Prior to 1963, virtually all women with phenylketonuria (PKU) who were of childbearing age were mentally retarded and bore few, if any, children. The introduction of the Guthrie test, a simple, rapid, and economical method of screening neonates for elevated levels of blood phenylalanine, in 1963, however, made it possible to initiate early treatment of affected children with restricted levels of dietary phenylalanine. Such dietary treatment resulted in normal physical and intellectual development among children with PKU.

In 1980, Lenke and Levy demonstrated an association between blood phenylalanine levels during pregnancy and the prevalence of mental retardation, microcephaly,

congenital heart disease, spontaneous abortion and low birth weight in the offspring of PKU women. This discovery led to the prediction that the incidence of new cases of PKU-related mental retardation would return to its former level within one generation, if women with PKU reproduced at a normal rate and were without dietary restriction of phenylalanine during or before pregnancy. In response to this public health issue, the NICHD initiated a collaborative study designed to evaluate the efficacy of a phenylalanine-restricted diet during or before pregnancy in reducing the fetal morbidity associated with MPKU. Participants came from the major PKU treatment centers in the United States and Canada, and several centers in Germany.

Among the 575 pregnancies enrolled in the study, 413 resulted in live births, while 76 spontaneously aborted, 80 had elective termination, three were stillbirths, and three were ectopic pregnancies. Of the 572 pregnancies that were followed, phenylalanine-restricted diet was initiated before conception in 147 women (26 percent) and in 207 women (36 percent) by eight gestational weeks. However, only 15.9 percent of pregnancies that resulted in live births maintained phenylalanine levels below 10 mg/dL from before conception, and throughout the entire pregnancy to birth. Another 18.4 percent established this level of control by the first 10 gestational weeks. When compared with the results of Lenke and Levy (1980), dietary treatment clearly improved pregnancy outcome for many of these women. Microcephaly declined from the 73 percent reported by Lenke and Levy to 23 percent. When only those pregnancies with dietary control before conception were evaluated, the rate of microcephaly approached that of the normal population (3.6 percent). Congenital heart disease was not recorded in pregnancies of women treated before conception or in those in control by eight gestational weeks. Congenital heart disease was identified and confirmed via postnatal echocardiography or at autopsy in 34 offspring from pregnancies in women with PKU and in one offspring from a woman with mild hyperphenylalaninemia. These offspring included 33 live-born and two fetuses from pregnancies that were terminated in the second trimester because the congenital heart disease observed was considered inoperable and incompatible with postnatal survival. Women who were in metabolic control before conception or by 10 gestational weeks had offspring whose McCarthy General Cognitive Index (GCI) scores exceeded the mothers' by six-to-10 points, comparable to the offspring of women with untreated mild hyperphenylalaninemia
(GCI = 99), and controls (GCI = 107). Offspring of mothers who did not establish control until 10-to-20 gestational weeks had scores nearly identical to those of their mothers. If a mother was not in metabolic control until after 20 gestational weeks, the mean offspring score was 10 points below that of the mothers. Preliminary data on the Wechsler Intelligence Scale for Children-Revised (WISC-R) scores at seven years of age tend to support these observations.

The recommended level of blood phenylalanine during pregnancy that resulted from this collaborative study is two-to-six mg/dL. Close cooperation between the attending obstetrician and a metabolic team experienced in the care of persons with PKU yields the best outcome.

FAMILY FUNCTIONING

Adjustment in families rearing children with developmental disabilities is a complex, multidimensional construct that involves parents, siblings, and children with developmental disabilities. These individuals interact in ever-changing ways with each other and the outside world, in a life-course that is far from static. In 1987, the prevailing view of these families was one of pathology and maladjustment. Over the past decade, however, studies have challenged this view through the observations that compare families who knowingly adopted children with disabilities with families who had similar children by birth. Although researchers expected adoptive parents to be better adjusted, especially early on, after the initial crises, birth families adjusted quite well to the challenges posed by rearing a child with disabilities. Few differences were obvious when the children were, on average six years of age, although long-term adjustment might be better in adoptive parents. When the children were 11, however, some differences reemerged. Depression had risen slightly, but significantly, for both adoptive and birth mothers, but more so for birth mothers. The most noticeable difference was observed between adoptive and birth mothers in a personality measure of mental stability/instability.

These families are currently experiencing what is likely to be a stressful time for them—their children's transition to adulthood. With the passage of their children into adulthood and adult services, parents must confront a complex and unorganized group of programs where they are not assured availability, accountability, or appropriateness. Such an increasing challenge is likely to increase differences in adjustment between the relatively more and less-stable parents. Stable and enduring factors, such as personality traits and religious beliefs, help to predict how well parents will cope with their children's transition to adulthood. Further, observing how adoptive and birth families interact in at-home situations helps better assess how family members affect one another.

Additional studies involving family interactions focus on stress in families of young children with various syndromes, including Down, Williams, and Smith-Magenis. Other families with children who are at risk both biologically and socio-environmentally, for impaired development of arousal and attention, particularly children exposed prenatally to stimulants like cocaine, have shown that these children are more susceptible to stress factors, and, therefore, are more vulnerable to detrimental effects.

Researchers are also examining facilitation of social outcomes in groups of school-aged children with mild-to-moderate mental retardation, including Down syndrome and non-Down syndrome children with learning disabilities. As with all children, the quality of family interaction influences children's social skills and behavioral adjustment, which in turn, affect peer relationships and other social outcomes. Direct family member efforts to arrange and monitor social encounters for the children help increase their social participation, thus providing experiences that improve social skills. For children with

disabilities, studies note that both family interactions and the manner in which family members help children cope with social rejection influenced the children's ability to process social information and solve social problems.

AUTISM

Autism is a developmental disability that is evident in infancy or during preschool period and affects some of the most essential human behaviors, including the ability to: communicate feelings and ideas, interact socially, and establish and maintain relationships with others. The autism spectrum disorders include classic autism or autistic disorder, Asperger syndrome, Rett syndrome, Childhood Disintegrative Disorder, and Pervasive Developmental Disorder - Not Otherwise Specified. International epidemiological studies conclude that classic autism occurs in one-in-1,000 children, while the broader spectrum of autistic disorders occur in one-in-500. Recent reports from US service providers indicate that even these prevalence estimates may not reflect the true extent of this public health problem that affects most of these children throughout their life spans. Autism appears to occur with equal frequency in all races and social classes but affects three-to-four times as many boys as girls. It may co-occur with other disorders, notably mental retardation, epilepsy, or obsessive-compulsive disorder. In addition to difficulties in verbal and non-verbal communication, problem behaviors such as tantrums, physical aggression against self and others, property destruction, and stereotypies may also be part of the clinical picture. At the present time, there is no cure for any of the autism spectrum disorders, but intensive behavioral interventions do dramatically alter the prognoses for children. Some pharmacological agents are also effective in reducing symptoms.

The NICHD leads the NIH in its support of autism-specific research, particularly that focused on neurobiology and genetics. The NICHD uses most of the available grant mechanisms available to support autism research. Infrastructure support is also provided for autism research through the MRRCs, in conjunction with grants funded directly by NICHD or in collaboration with other NIH Institutes. Many research topics concerning autism are currently under investigation:

- Genetic studies to map genes that are located on Chromosome 15 and their expression in autism.
- Developmental studies to evaluate developmental models based on central nervous system infection, expression of early developmental genes, environmental triggers, and neural tube injury.
- Neurochemical and neuropharmacological studies that focus on serotonin metabolism in the autistic brain as well as behavioral and biochemical mechanisms of self-injury.

- The development of methods in neuroimaging is augmenting and facilitating studies of stimulus overselectivity and restricted stimulus control, facial and emotional recognition, and behavioral and bio-behavioral aspects of abnormal stereotyped behaviors.
- Family studies complement other working research on joint attention, symbolic behavior, and prosody and pragmatics in communication in autism.
- Studies of developmental behavioral pharmacology have led to further studies involving clinical trials of behavioral, bio-behavioral, and biological treatments, some of which include rational drug design.
- Research, funded in part through an innovative small-business grant mechanism, has helped to develop computer- and virtual reality-assisted academic, communication, and social instruction.
- Professionals and lay readers can obtain more information on this research at the NICHD autism Web site at http://www.nichd.nih.gov/autism.

NETWORK ON THE NEUROBIOLOGY AND GENETICS OF AUTISM: COLLABORATIVE PROGRAMS OF EXCELLENCE IN AUTISM (CPEAs)

In 1997, in response to the recommendations of the *Autism: State of the Science Conference* held in 1995 and through an RFA, the NICHD, with co-funding from the National Institute on Deafness and Other Communication Disorders, established the Network on the Neurobiology and Genetics of Autism, referred to as the CPEAs (Figure 5). Each multidisciplinary, often multi-site project is studying some particular basic and clinical aspect of the biological etiology (including possible genetic, immunological, and/or environmental causes), brain structure and function, and clinical course of autism. Each multidisciplinary project has a unique focus and research plan; in addition, all projects use a common diagnostic protocol and common core measures and procedures to collectively address some research questions that are beyond the resources and/or subjects of any single project. Individually or collectively, the CPEAs investigate the causes, diagnosis, early detection, prevention, and treatment of autism. Expertise at the CPEAs ranges from immunology, molecular genetics, and developmental biology, to clinical and developmental pharmacology. The Network also participates in an international autism genetics research consortium that pursues autism research of international import.

Coordination of information across CPEAs within and, as appropriate, outside the Network occurs through regular meetings of the Network Steering Committee. These annual scientific meetings include investigators from all projects and ongoing subcommittee working groups who discuss topics such as genetics, cognitive development, and communication, supplemented with e-mail and telephone conferencing among the directors of the CPEAs. Extensive outreach and sharing of strategies for recruitment and retention make individuals aware of opportunities to participate in the

CPEA research projects. The more than 1,600 probands who are currently available for study will ultimately increase to more than 4,500, well-characterized research participants for whom genetic and phenotypic data were gathered from both multiplex and singleton families. Both competitive and administrative supplements to these projects have allowed NICHD to take advantage of this shared resource in facilitating the development of methods in genetic analysis, neuroimaging (e.g., correction of motion errors), neuropsychology (CANTAB-automated neuropsychology battery), and in the conduct of clinical research studies on important questions regarding public health.

In the first three years of the Network's existence, individual CPEA-specific objectives that were carried out yielded more than 130 publications (plus abstracts). These projects have developed a core of common biological and behavioral measures, provided staff members across the Network with training on the reliability of the measures, and led to six major trans-Network projects now in progress. Such projects include collaborative studies on the candidate genes *HoxA* and *HoxB*, language and communication, executive function, head circumference, early cognitive development, treatment with secretin, and a 12-site, case-control study of regression and vaccination in 1,600 persons with autism and 1,250 healthy controls.

Individually, the CPEAs have also generated a number of important findings. Investigators at the University of Washington have identified potential early behavioral (e.g., social orienting responses) and biological markers for autism (e.g., ERP brain responses to familiar versus stranger's face). A collaboration of investigators at the University of Washington, the University of Chicago, and Yale University has validated the concept of autism regression starting around two years of age. In large numbers of these cases, however, closer study has provided evidence across projects of actual indices of developmental problems much earlier in infancy.

Investigators at the University of Washington have also found that very young children with autism show an auditory preference for asocial mechanical signals, rather than the cross-cultural normal preference for simplified caregiver speech called "motherese." Functional brain imaging studies done at Yale University have led investigators to suggest that children with autism process social information differently from normal healthy controls, using for social processing that region of the brain normally used for processing information about objects for social processing.

Macrocephaly, a head circumference above 97th percentile for age and sex, is found almost three times more often in autism spectrum subjects than in controls. When investigators at the University of Utah correlated head size with differential performance, they found evidence that a brain mechanism that results in enlargement of the brain and head may be involved in the pathogenesis of autism, for at least a significant subset of patients. Children with autism consistently have significantly larger brains when

compared to healthy controls in data collected across projects at the University of Washington, the University of Utah, and the University of California at Irvine. Further, investigators at the University of Washington have found that abnormal structural/chemical brain findings are related to severity of symptom expression in autism.

Of the children with autism who were examined at the University of Utah, 50 percent show immune associations, such as associated human leukocyte antigen (HLA) haplotypes, deficient complement protein C4B, or other immune indicators. Ongoing research is identifying particular HLA haplotypes that appear to be associated with autism and haplotypes that appear to be protective from autism.

Conflicting evidence for a core deficit in acquisition and performance of executive function has led investigators at the University of Utah to suggest that persons with autism may perform above expectations on measures of executive function administered via computer, versus those administered by a person in a social situation. Further, investigators at the University of Pittsburgh and Carnegie Mellon University have obtained increasing evidence of a profile or pattern of impaired and intact abilities in adults with autism. This impairment is indicative of a selective disorder of complex information processing, without attendant overall deficits in verbal working memory on particular tasks. In their studies of subcortical and cerebellar regulation of sensorimotor abilities, these investigators have found that those abilities subserving visually guided saccadic eye movement remain intact.

Investigators involved in imaging studies of autistic brains have developed and implemented a new method for correcting functional imaging data for the effects of heartbeat and respiration. This methodology is accompanied by the development of motion correlation algorithms that exceed the accuracy of existing methodologies. Investigators at the University of Pittsburgh have developed new statistical methods for analyzing functional imaging data across time, as well as more sensitive methods for analyzing autistic differences on fMRI.

PRENATAL DIAGNOSIS

The NICHD has a long history of support for research on prenatal diagnosis. In 1971, the Institute initiated a collaborative study that supported nine institutions across the US to obtain scientifically valid information on mid-trimester amniocentesis for the prenatal diagnosis of genetic disorders. Mid-trimester amniocentesis is now considered a highly accurate and safe procedure that does not significantly increase risk to the mother or the fetus. Based on another study that was initiated in 1985, eight centers concluded that first-trimester chorionic villus sampling (CVS) is a safe and effective technique for the early prenatal diagnosis of cytogenetic abnormalities. CVS, however, entails a slightly higher risk of procedure failure and of fetal loss when compared to amniocentesis. A subsequent randomized clinical comparison of transcervical (TC) and transabdominal (TA) CVS conducted by the same group of investigators concluded that TC and TA CVS are comparably safe and accurate procedures for diagnosis of chromosomal disorders during the first trimester.

EARLY AMNIOCENTESIS VS. LATE TRANSBDOMINAL CVS (THE EATA TRIAL)

Following completion of the second study on TC/TA CVS, the investigator group proposed a further trial to compare the safety and efficacy of CVS with early gestational performance of amniocentesis in the same gestational period. The project initially aimed to compare early amniocentesis to transabdominal CVS performed from 11 through 14 weeks gestation. Almost immediately problems were observed in the earliest gestational week with occurrence of club foot deformities in infants from a small cohort of mothers. Soon after that, relatively compelling data from this trial and a Canadian study made participating obstetricians anxious, prompting the safety committee to stop recruitment during the 12^{th} gestational week. When similar observations were made in a Danish study, the Center's advisors recommended dropping the 11^{th} gestational week from the eligibility criteria.

The EATA trial has slowly, but steadily recruited over 3,000 patients at the 13^{th} and 14^{th} gestational weeks. Clubfoot deformity was found in slightly over 1.5 percent of infants sampled during the early weeks (compared to approximately 0.5 percent from background), while the percentage was less significantly elevated in those sampled at the 13^{th} gestational week.

As an outgrowth of these projects and a direct result of the media focus on a few reports of limb reduction defects (LRD) following CVS, the NICHD has indirectly supported an ongoing data collection on this particular issue. The data are collected in a prospective manner under the continuing identity of the CVS Registry, originally supported by

minimal funds from the World Heath Organization (WHO). This project was continued as a form of unbiased sequential data collection to allow reasonable comparison with similar clinical data collected from birth defect registries in existence in several parts of the world, including the long-standing British Columbia Birth Defect Registry. The WHO registry now contains data on approximately 220,000 consecutive CVS cases with an LRD incidence of about 4.5-in-10,000 births or equivalent to the worldwide population of LRD as recorded in multiple birth defect registries. When CVS is performed at six-to-seven gestational weeks for religious preference in abortion decisions, increased incidence of LRD is observed. Women should avoid these procedures unless specific counseling and warnings are given, where the patient's preference overrides these warnings. In standard clinical performance of CVS, the risk is not increased for subsequent LRD occurrence.

Although mid-trimester amniocentesis and first-trimester CVS are considered accurate and relatively safe procedures, these invasive techniques carry some, though minimal, risks to the mother and the fetus. For this reason, NICHD decided to publish a request for contract proposals for a collaborative project, with the main objective of developing a non-invasive, safe, relatively inexpensive and accurate technique for the prenatal diagnosis of genetic disorders that doctors can perform during the first trimester.

THE NICHD FETAL CELL STUDY (NIFTY)

NIFTY is a prospective multi-center study designed to determine the sensitivity, specificity, and predictive value of using fetal cells isolated from maternal blood in detecting fetal gender and fetal chromosomal abnormalities, when compared to the results of amniocentesis, CVS, or the outcome of pregnancy. Pregnant women who are seeking prenatal diagnosis using either amniocentesis or CVS are eligible to participate in this on-going study if they are 16 years of age or older, with an assigned gestational age between 10 weeks/0 days and 20 weeks/6 days; candidates must also be at high risk for fetal aneuploidy because they are 35 years of age or older at estimated date of delivery, and/or at elevated risk for any or several of the following: fetal sonographic abnormality, prior history of fetus with aneuploidy, abnormal maternal serum screen, and/or comparable risk as determined by the study physician. Fetal nucleated red blood cells are separated from maternal blood by fluorescent activated cell sorting or magnetic activated cell sorting. Enriched cells are then evaluated by FISH using DNA-specific probes for Chromosomes X, Y, 13, 18, and 21. Cytogenetic confirmation is accomplished by comparing the FISH results to those of amniocentesis, CVS, or pregnancy outcome.

Current NIFTY data indicate overall detection rates of 47 percent for autosomal aneuploidy and 53 percent for sex chromosome aneuploidy, rates that are comparable to *single* parameter antenatal screening tests. Magnetic sorting yields superior results with an overall detection rate as high as 72 percent in a small series. Analysis of the effects of delayed sample processing, as might occur when samples are shipped to a central laboratory, has shown that the delay actually improves the detection rate. Thus, although the technology is not yet applicable to clinical practice, the study is indicating productive avenues for significant improvement in aneuploidy detection.

In December 1999, the scope of the study was expanded to detect single-gene defects, in addition to aneuploidy, using intact cells or plasma. The use of the plasma portion of the maternal blood sample has proved especially fruitful. Correlations between levels of fetal DNA in the maternal plasma appear predictive of early maternal pre-eclampsia and may even indicate an increase in risk for some autosomal trisomies (specifically, 21 and 13). Investigators also found that the detection of paternal RhD contribution is reliable and may soon eliminate the need for invasive testing or Rhogam® use in pregnancies of heterozygous fathers, where the "D" allele is contributed to the fetus; this test prevents fetal mortality and maternal complication. There is great potential in applying this form of non-invasive prenatal diagnosis to any condition in which the paternal genetic contribution is the deciding factor in the anticipated fetal disease risk. Thus, doctors can diagnose cytogenetic and autosomal dominant genetic conditions that stem from the paternal contribution, using non-invasive analysis of free fetal DNA in circulating maternal plasma. Similarly, 50 percent of autosomal recessive conditions caused by compound heterozygous parental contributions (different maternal and paternal mutations) can be ruled out non-invasively. More recently, free fetal RNA was also found in the maternal plasma, opening up an entirely new area of application to non-invasive prenatal diagnosis that is not limited to paternal genetic contributions. In addition, investigators are developing culture techniques in an effort to expand fetal cell number.

FIRST AND SECOND TRIMESTER EVALUATION OF ANEUPLOIDY RISK (FASTER)

FASTER is a multi-center prospective study that compares first- and second-trimester non-invasive methods of screening for fetal aneuploidies with currently accepted, standard-of-care programs of second trimester maternal serum screening. First-trimester screening, taken together with maternal age, involves an ultrasound measurement of fetal nuchal translucency thickness at 10-to-14 gestational weeks, as well as serum levels of pregnancy-associated protein-A and free β human chorionic gonadotropin.
Second-trimester screening is based on the current, standard-of-care, serum "triple screen," which consists of measurement of levels of alpha fetoprotein, unconjugated estriol, and human chorionic gonadotrophin (hCG), performed at 15-to-18 gestational

weeks, taken together with maternal age and serum inhibin-A levels (so called "quad test"). A consortium of 14 clinical centers will recruit an estimated 62,000 patients, a sample size based on an 8 percent advanced maternal age and an anticipated attrition rate of 20 percent. To date, 22 percent of the sample are 35 years and older, with an attrition rate of 7 percent. A smaller sample size should provide the target goal of 100 Down syndrome births. Each patient will have both first-and second-trimester methods of screening performed. To date, 12 cases of Down syndrome, three cases of Trisomy 18, and one case of Trisomy 13 were born in the population of FASTER enrollees.

Preliminary analysis of data shows that first-trimester screening protocol currently has a detection rate of 80 percent, with a 2.2 percent screen-positive rate, compared to the projected detection rate of 70 percent and screen-positive rate of 2.0 percent. The second-trimester screening has yielded a detection rate of 84 percent, with a screen-positive rate of 7.2 percent, compared to the anticipated detection rate of 75 percent and a 5.0 percent screen-positive rate. Taken together, the overall first-and second-trimester screening program provides a 95 percent detection rate for Down syndrome, with a 9.0 percent screen-positive rate.

Practitioners in the US tend to incorporate first-trimester screening into standard clinical practice. Many commercial organizations are now becoming involved in this area, offering first-trimester screening services to patients as a commercial enterprise. In October 1999, the American College of Obstetricians and Gynecologists issued a position statement that such screening is investigational and should not be used in routine clinical practice. Because it is the only study that can adequately answer the question of whether first-trimester screening is better or worse than the current, standard-of-care, second-trimester screening, the results of FASTER are of vast significance.

APPENDIX 1: MRDD BRANCH ACTIVITIES

CONFERENCES AND WORKSHOPS

- *Fetal Therapy*—April 17-18, 1998
- *Dendritic Abnormalities in Mental Retardation and Developmental Disabilities*—April 30-May 1, 1998
- *Nobel Minisymposium on Fetal Medicine*—May 28-29, 1998
- *NIH State of the Science in Autism: Screening and Diagnosis Working Conference,* Jointly sponsored with the NIMH, NINDS, and NIDCD—June 15-17, 1998
- *Genetics of Rett Syndrome: Research Strategies*—June 16-17, 1998
- Working meeting on *Secretin Treatment Protocol Conference*, Sponsored by ORD—December 1998
- Workshop on *Fragile X Syndrome: Future Research Directions*—December 3-4, 1998
- Parent and the Child's World: Multiple Influences on Intellectual and Social-Emotional Development, Jointly sponsored with Robert Wood Johnson Foundation—August 2-3, 1999
- Annual Conference of the Network on the Neurobiology and Genetics of Autism: Collaborative Programs of Excellence in Autism—September 29, 1999
- Treatments for People with Autism and Other Pervasive Developmental Disorders: Research Perspectives, jointly sponsored with the NIMH, NINDS, and NIDCD—November 8-9, 1999
- Etiology and Pathophysiology of Self-Injurious Behavior: Implication for Treatment—December 5-6, 1999
- Workshop on Early Childhood Neurobehavioral Assessment Tools in Response to Teratogenic Effects, Jointly sponsored with NIAAA, NINDS, and NIEHS—March 8-10, 2000
- Annual NIH Autism Coordinating Committee and Autism Research Advocacy Group Meeting, Jointly sponsored with the NIMH, NINDS, and NIDCD—March 14, 2000
- International Genetics of Autism Meeting—March 17-18, 2000
- *Neurofibromatosis Workshop*, Jointly sponsored with NINDS—May 4-5, 2000
- *ICF/Rett Syndrome Meeting*, Jointly sponsored with NINDS and the International Rett Syndrome Association—June 13-15, 2000
- *XXY/Klinefelter Syndrome: Expanding the Phenotype and Identifying New Research Directions*—August 28-29, 2000

- *The Olfactory Model System and Rett and Kallman Syndromes: Sniffing Out Insights into Brain Development*, Jointly sponsored with ORD, NIDCD, NIMH, and NINDS—September 12, 2000
- *Invasive Fetal Diagnosis and Therapy in the Third Millennium*—September 22-23, 2000
- *Annual Conference of the Network on the Neurobiology and Genetics of Autism: Collaborative Programs of Excellence in Autism*—September 25-27, 2000
- *Consensus Development Conference on Phenylketonuria (PKU): Screening and Management*—October 16-18, 2000.
- *Autism Regression/Vaccination Study Protocol Planning* Meeting—November 6, 2000

FUTURE PLANS

- Workshop on Chromosome 18: A "State-of-the Science" meeting, to be convened in the spring of 2001, will bring together basic and clinical investigators familiar with syndromes associated with Chromosome 18, as well as basic and clinical investigators with expertise from other areas of research with direct relevance to disorders of Chromosome 18. This meeting will establish a consensus of the state of research in the study of disorders associated with Chromosome 18. It will also identify critical areas of research where fundamental knowledge relevant to the studies of these disorders is lacking, areas that could be the focus of study by investigators through existing funding mechanisms at the NIH.
- International Conference on Maternal Phenylketonuria (MPKU)
- Conference on Fetal Therapy
- Workshop on Gene Therapy
- Fragile X Conference
- RFA on Self-Injurious Behavior

APPENDIX 2: MRDD BRANCH PERSONNEL

Felix de la Cruz, M.D., M.P.H., a pediatrician, was appointed Chief of the MRDD Branch in 1989. He is responsible for the clinical and biomedical aspects of mental retardation and developmental disabilities, including inborn errors of metabolism, malnutrition, prenatal diagnosis, fetal therapy, and clinical genetics.

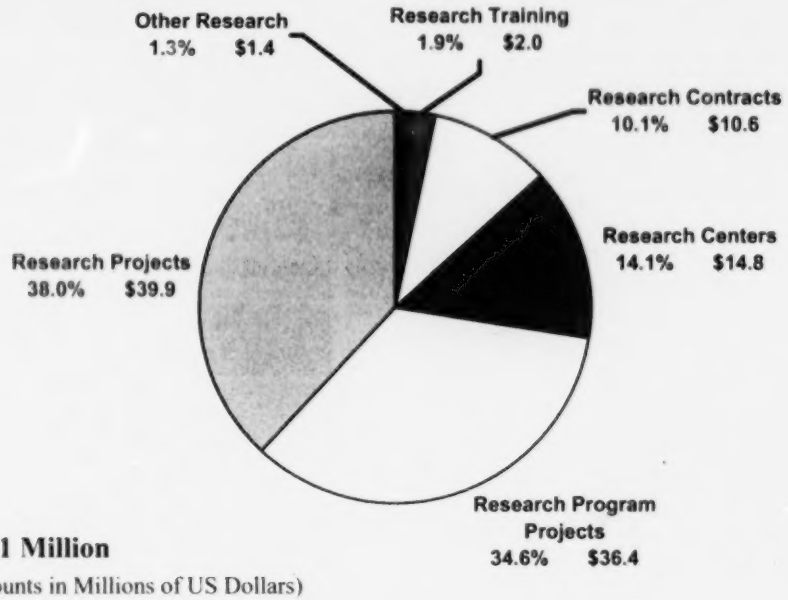
Marie Bristol-Power, Ph.D., is an educational psychologist who joined the MRDD Branch in 1995. She currently serves as the Special Assistant on Autism in the Office of the Director of the NICHD. Responsibilities for the behavioral and bio-behavioral research in mental retardation and developmental disabilities, including cognitive neuroscience, social and emotional behavior, family research, behavioral intervention, and psychopharmacology that Dr. Bristol-Power previously handled are now undertaken by Drs. de la Cruz and Oster-Granite.

Mary Lou Oster-Granite, Ph.D., is a developmental neuroembryologist, neurovirologist, and neurogeneticist. A long-time grantee of the NICHD, she joined the MRDD Branch in 1999. Dr. Oster-Granite formerly served as a tenured Professor of Biomedical Sciences at the University of California at Riverside. Her research interests have included genetic animal models of mental retardation and developmental disabilities, most recently, animal models of Down syndrome and of ornithine carbomyl transcarbomylase deficiency. Her programmatic interests include brain function at a cellular/molecular level, developmental neurobiology, neurochemistry, neurovirology, molecular genetics, and gene therapy.

Ms. Barbara Alvarado, Program Assistant, is an invaluable new addition to the MRDD Branch. She coordinates all the programmatic and administrative activities within the Branch and, in performing those responsibilities, plays an indispensable role in managing the daily activities of the Branch.

FIGURES AND TABLES

Figure 1: MRDD Branch Distribution of Funds by Support Mechanisms for Fiscal Year 2000



Total: $105.1 Million

(All Dollar Amounts in Millions of US Dollars)

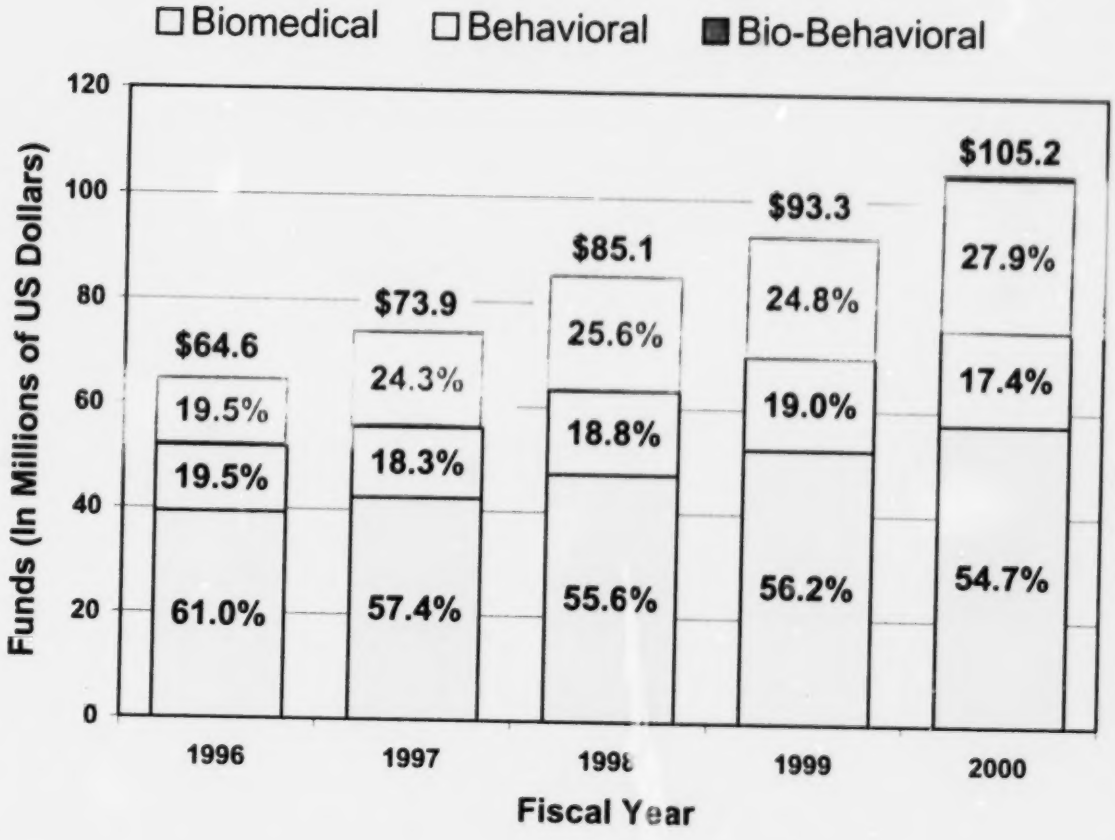
Table 1: MRDD Branch Projects Funded by Support Mechanisms for Fiscal Year 2000

Support Mechanisms	Number of Projects	Funds
Research Projects* (excluding R01s and P01s)	45	$ 6,747,944
R01s	100	33,163,956
P01s	34	36,392,727
Program Research Centers	14	14,816,038
Career Awards	13	1,285,827
National Research Service Awards	13	1,977,600
Research Contracts	11	10,593,756
Other Research**	11	138,318
Totals	**241**	**$105,116,166**

* R03, R15, R21, R29, R37, R41, R43, R44

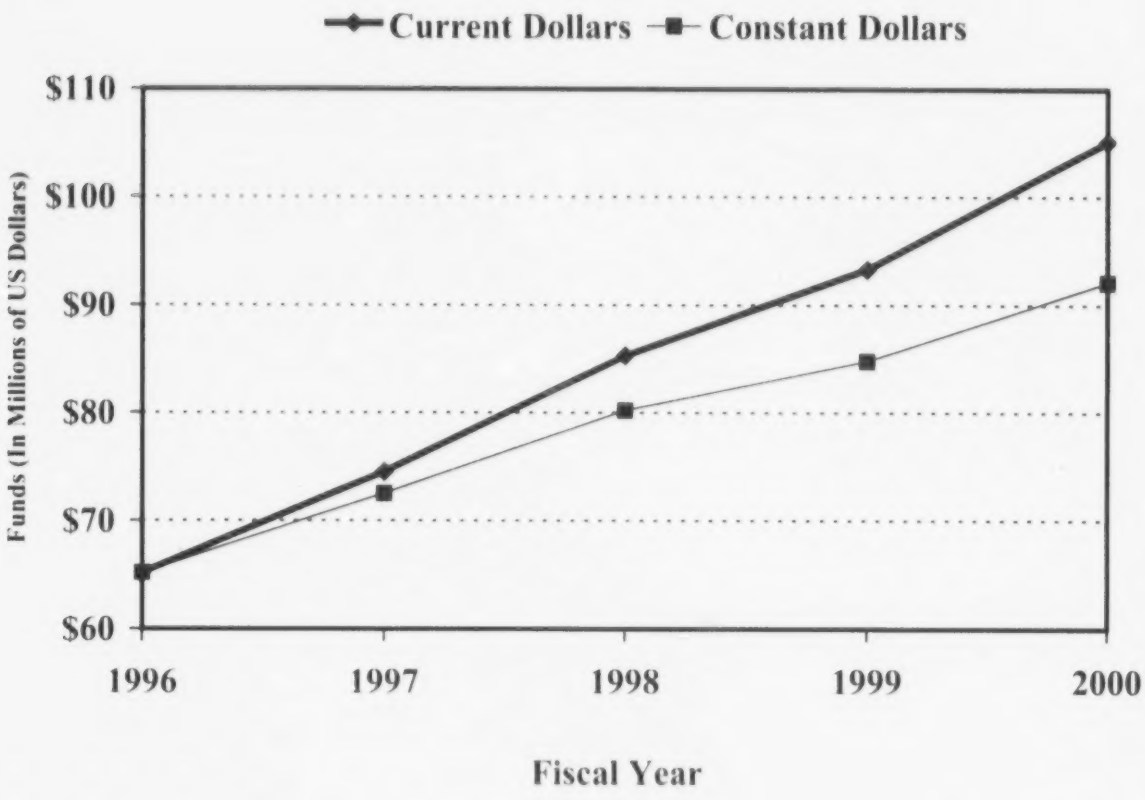
** R13, T15

Figure 2: MRDD Branch Biomedical, Behavioral, and Bio-Behavioral Projects for Fiscal Years 1996–2000



BEST COPY AVAILABLE

Figure 3: MRDD Branch Funds in Current and Constant Dollars for Fiscal Years 1996–2000



Base Year (1996 = 100)

Figure 4: The NICHD Mental Retardation Research Centers (MRRCs)

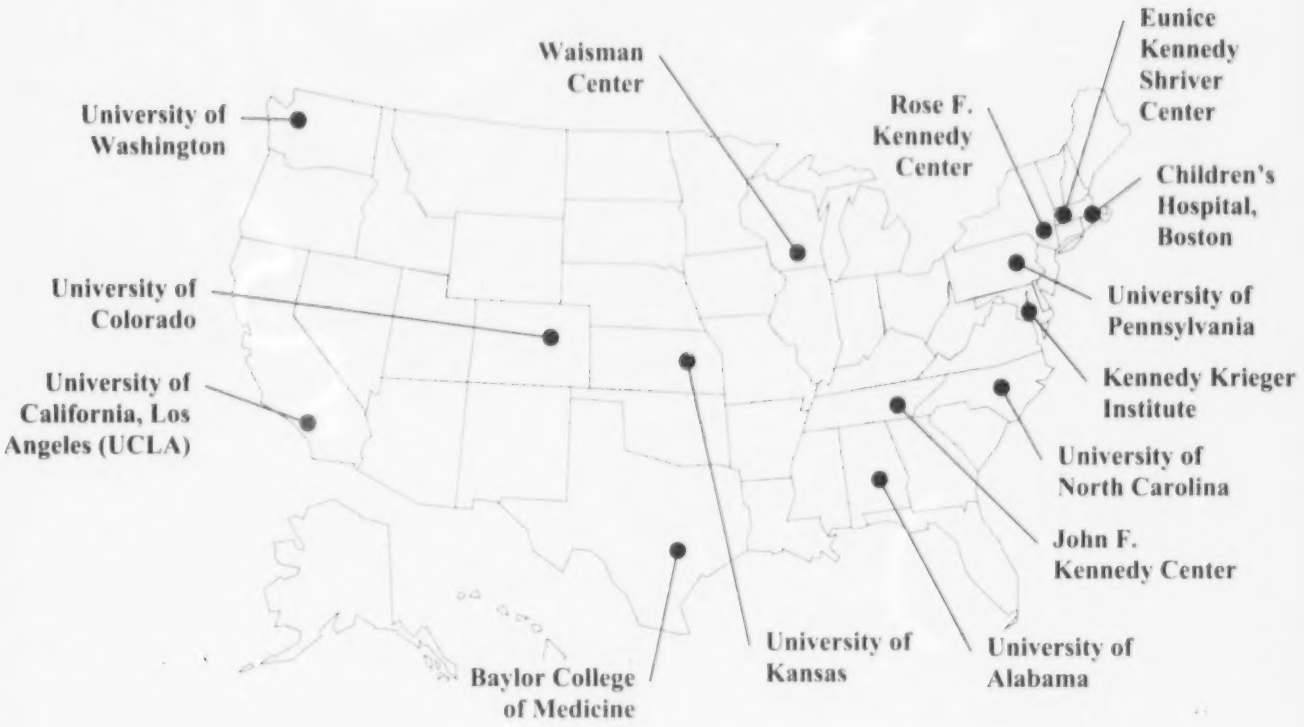


Figure 5: The NICHD Collaborative Programs of Excellence in Autism (CPEAs)

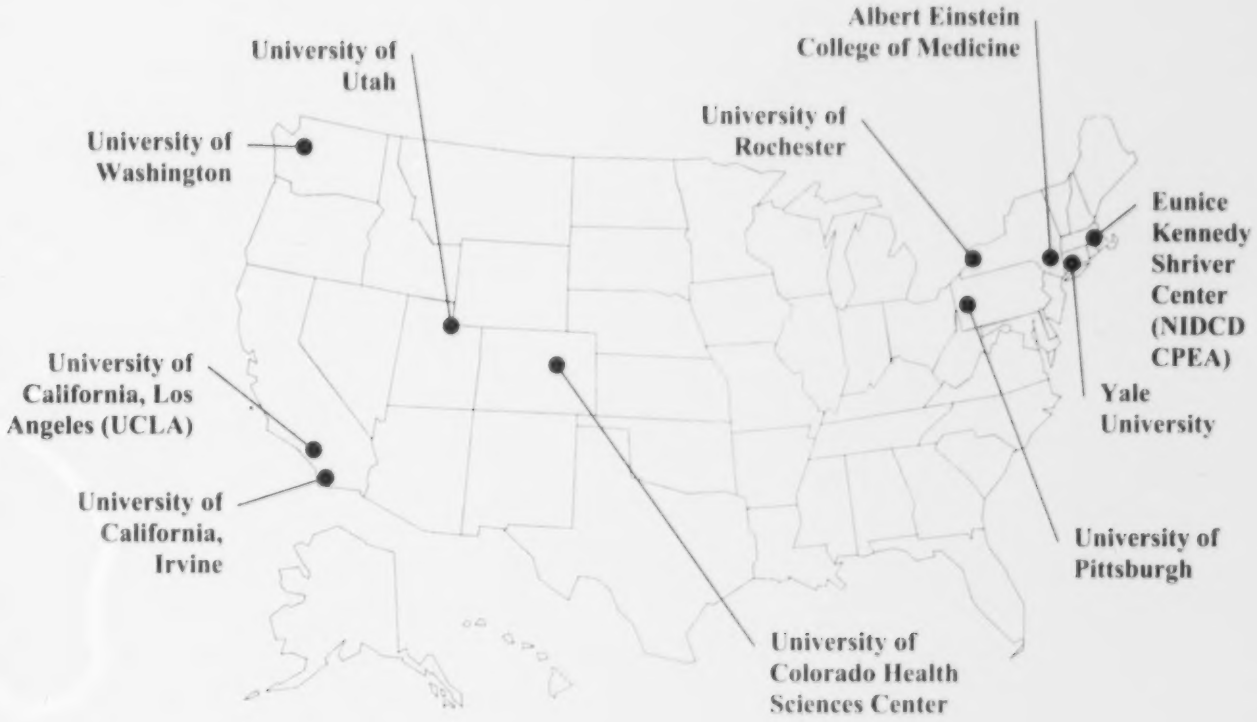

Albert Einstein College of Medicine
University of Utah
University of Rochester
University of Washington
Eunice Kennedy Shriver Center (NIDCD CPEA)
University of California, Los Angeles (UCLA)
Yale University
University of California, Irvine
University of Pittsburgh
University of Colorado Health Sciences Center

Table 2: Diseases and Syndromes—MRDD Branch Research Support for Fiscal Year 2000

Adenylsuccinase Deficiency
Adrenoleukodystrophy
Aicardi Syndrome
AIDS Dementia
Alpha-1-Antitrypsin Deficiency
Angelman Syndrome
Arginase Deficiency
Asperger Syndrome
Autism
Batten Disease
Bilirubinemia
Canavan Disease
Cerebral Hemorrhage (VLBW Infants)
Cerebral Palsy
Charcot-Marie-Tooth Syndrome
Chromosome 18q Syndrome
Cohen-Van Dyke Syndrome
Cri du Chat Syndrome
Craniosynostosis
Cystic Fibrosis
Di George Syndrome
Down Syndrome
Duchenne/Becker Muscular Dystrophy
Epilepsy
Fetal Alcohol Syndrome
Fragile X Syndrome
Fucosidosis (Glycolipid Storage)
Galactosemia
Gangliosidoses
Gaucher Disease
Glutaric Acidemia
Goltz Syndrome
Gyrate Atrophy
Holoprosencephaly
Homocystinuria
Hurler Syndrome
Hyperpipecolic Acidemia
Hypothyroidism
Hypoxic-Ischemic Encephalopathy
Isotretinoin Embryopathy
Krabbe Disease
Lead Toxicity
Leigh Syndrome
Lesch-Nyhan Syndrome
Lissencephaly
Malnutrition
Maple Syrup Urine Disease
Mannosidosis
Maternal Phenylketonuria (MPKU)
MELAS Syndrome
MERRF Syndrome
Mucopolysaccharidosis Type VII
Neural Tube Defects
Niemann-Pick Disease Type A
Organic Acidemias
Ornithine Transcarbamylase Deficiency
Peroxisomal Disorders
Phenylketonuria (PKU)
Prader-Willi Syndrome
Propionic Acidemia
Pyruvate Carboxylase Deficiency
Radiation Injury
Refsum Disease
Rett Syndrome
Rhizomelic Chondrodysplasia Punctata
Retinitis Pigmentosa
Smith-Magenis Syndrome
Smith-Lemli-Opitz Syndrome
Spinocerebellar Ataxia
Tardive Dyskinesia
Tourette Syndrome
Turner's Syndrome
Treacher Collins Syndrome
Velocardiofacial Syndrome
Von Recklinghausen's
Williams-Beuren Syndrome
WAGR Syndrome (Wilms Tumor)
X-Linked Disorders
Zellweger Syndrome